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THE IMPACT OF PROCESSING METHODS ON THE NUTRITIONAL AND PHYSICOCHEMICAL PROPERTIES, AND SCREENING PHOTOCHEMICAL COMPOSITIONS OF A MUCUNA PRURIENS SEED

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ABSTRACT

A phytochemical analysis was conducted on a M. prunes seed extract to determine the presence of various bioactive compounds. Qualitative tests were performed to detect alkaloids, carbohydrates, reducing sugars, glycosides, cardiac glycosides, proteins and amino acids, flavonoids, phenolic compounds, tannins, phlobatannins, saponins, phytosterols, cholesterol, terpenoids, triterpenoids, anthraquinones, anthocyanins, carboxylic acids, and resins. The results revealed the presence of several phytochemical classes, suggesting potential medicinal properties of the plant extract. This study also investigated the effects of different processing methods – boiling, roasting, and soaking – on the moisture content, protein content, crude fiber, crude fat, carbohydrate content, pH, and ash content of a specific food matrix. Raw samples were compared to processed samples to determine the changes induced by each treatment. The results indicate that processing significantly affects the nutritional composition and some physicochemical properties. Roasting led to a substantial reduction in moisture content (range: 6.21-7.29%) and an increase in crude fat (range: 4.48-4.91%), while boiling (moisture range: 10.24-11.93%; protein range: 21.05-23.42%; ash range: 2.98-4.24%) and soaking (moisture range: 10.97-11.92%; protein range: 21.37-22.50%; ash range: 2.98-3.40%) resulted in higher carbohydrate content (boiled range: 66.46-67.92%; soaked range: 67.08-68.53%) and a decrease in protein and ash. The pH remained relatively stable across all processing methods (range: 6.4-6.5). These findings highlight the importance of processing techniques in modifying the nutritional profile of the food matrix, which has implications for its utilization and nutritional value.

KEYWORDS: Processing methods, moisture content, protein, crude fiber, crude fat, carbohydrate, pH, ash content, and screening.

1. INTRODUCTION

Food processing is an essential step in transforming raw agricultural commodities into palatable and safe food products, often leading to significant alterations in their chemical and physical characteristics, consequently impacting their nutritional attributes and overall quality (Fellows, 2017). Techniques such as boiling, roasting, and soaking are widely employed for various purposes, including enzyme inactivation, reduction of anti-nutritional factors, enhancement of sensory properties, and extension of shelf life (Rahman, 2009). Understanding the specific effects of these processes on the fundamental macronutrient and mineral composition of food is crucial for optimizing dietary intake and

informing food product development (Institute of Medicine, 2005). The current study concerned the proceesing of Mucuna Prunes seed. Mucuna seeds, also known as velvet beans, possess a wide range of physicochemical properties that make them a subject of interest for various industries. The variability in size, shape, and color of these seeds provides valuable insights into their composition and potential applications. Notably, mucuna seeds are rich in protein, with reported values ranging from 23% to 37% (Singh et al., 2017), and they also contain a significant amount of oil, with yields approaching 8% (Nwokocha et al., 2012). These high protein and oil contents make muccuna seeds an appealing option for both food and industrial purposes.

Mucuna pruriens seeds are a rich source of protein, carbohydrates, and minerals, including calcium, iron, and zinc. They also contain bioactive compounds such as L-DOPA, serotonin, and flavonoids, which have been shown to have various health benefits. Several studies have reported on the nutritional composition of Mucuna pruriens seeds, including their protein content, amino acid profile, carbohydrate content, and mineral composition.[1]

The functional properties of Mucuna pruriens seeds are important for their potential use as a food ingredient. Several studies have investigated the functional properties of Mucuna pruriens seeds, including their water absorption capacity, oil absorption capacity, emulsifying properties, and foaming properties.[2] These properties are important for the development of food products such as bakery products, meat products, and beverages.

Moreover, the presence of minerals and bioactive compounds such as phytic acid and tannins further enhances the potential value of mucuna seeds (Adebowale et al., 2005). This diverse range of nutrients and compounds in the seeds suggests potential applications in food, pharmaceuticals, and other industries. Therefore, a comprehensive characterization of mucuna seeds is essential to fully comprehend their composition and explore their potential uses in different sectors. This could involve detailed analysis of their nutritional content, functional properties, and potential applications in various products.

The physicochemical properties of Mucuna pruriens seeds make them a promising ingredient for the development of functional foods and nutraceuticals. Several studies have investigated the potential applications of Mucuna pruriens seeds, including their use as a protein source, as a functional ingredient in bakery products, and as a source of bioactive compounds for the treatment of various diseases.[3]

In conclusion, the physicochemical analysis of Mucuna pruriens seeds is an important tool for characterizing their nutritional composition, functional properties, and potential applications. The seeds are a rich source of protein, carbohydrates, and minerals, and contain bioactive compounds with various health benefits. The functional properties of the seeds make them a promising ingredient for the development of functional foods and nutraceuticals. Further research is needed to fully understand the potential applications of Mucuna pruriens seeds and to develop new products that can benefit from their unique properties.

1.1 General Objective of study

The main objective of this study is to determine Impact of Processing Methods on the Nutritional and Physicochemical Properties and Screening photochemical compositions, of Mucuna prurience seeds in Bonga Kaffa zone, South West Ethiopia, and assess their potential for medicinal and industrial applications.

1.2 Specific objectives of study

1. To screen phytochemicals compositions on Mucuna pruriens extract, identifying the presence of various bioactive compounds.
2. To determine and compare the effects of different processing methods (boiling, roasting, and soaking) on the moisture content, protein content, crude fiber, crude fat, carbohydrate content, pH, and ash content of Mucuna pruriens seed.

2. Literature reviews

Several studies have investigated the proximate composition and anti-nutritional factors of Mucuna pruriens seed and the effect of processing methods on its nutritional quality. A study by Oluwajuyitan et al. (2020), found that raw Mucuna pruriens seed contains phenol, phytate, tannins, oxalate, and saponin. The study also investigated the effect of roasting, germination, and fermentation on the proximate composition and anti-nutritional factors of Mucuna pruriens seed. The results showed that roasting and germination reduced the levels of anti-nutritional factors and improved the nutritional quality of the seed.

Another study by Oyeyinka et al. (2020), investigated the nutritional properties of Mucuna pruriens seed powder and the effect of processing methods on its nutritional quality. The study found that the seed powder had high carbohydrate and protein content and appreciable levels of L-Dopa. The study also found that processing methods such as boiling, roasting, and milling had varying effects on the nutritional quality of the seed powder. A review article by (Adhikari et al. 2019), assessed the potential nutritive and medicinal properties of Mucuna pruriens seed. The review found that the seed is a good source of protein, carbohydrates, and minerals and has potential health benefits, including improving male fertility and reducing symptoms of Parkinson's disease.

The study conducted (Ezegbe C. *et al.,* 2023), investigated the effects of various processing methods on the proximate composition and anti-nutritional factors in

Mucuna pruriens (velvet bean) seed flour. It also reveals that Anti-nutritional factors studied included phenol, phytate, tannins, oxalate, saponin, hydrogen cyanide, trypsin inhibitor activity and L-DOPA. Results showed that germination and fermentation increased crude protein and ash while other single treatments reduced nutrients.

The study conducted by Shanmugavel G. and Krishnamoorthy G. -2018, analyzed the nutritional and phytochemical properties of Mucuna pruriens seeds. This study reveals that the proximate analysis found the seed flour had high carbohydrate (54.1%) and energy (327 Kcal/100g) content and phytochemical analysis identified the presence of alkaloids, flavonoids, glycosides, saponins, steroids, tannins and terpenoids in muccuna purine seed. And it also concluded tha M. pruriens seeds are a rich source of nutrients and phytochemicals that could be utilized as therapeutic agents.

Overall, these studies suggest that Mucuna pruriens seed has potential as a food and feed crop due to its nutritional composition and potential health benefits. Processing methods such as roasting and germination can reduce anti-nutritional factors and improve the nutritional quality of the seed. Further research is needed to fully understand the nutritional properties and potential health benefits of Mucuna pruriens seed.

The legume family, Fabaceae, is the third largest among flowering plants, consisting of approximately 650 genera and 20,000 species (Doyle, 1994) and is the second most important plant source of human and animal nutrition (Vietmeyer, 1986). Some legume seeds are known for anti-cancerous compounds that retard or arrest the cancer growth. For instance, an alkaloid 'genistein' derived from kudzu beans (Pueraria Montana Lour.) has the unique property to retard cancer growth (Brink, 1995) and 'trigonelline' of jackbean (Canavalia ensiformis) possesses anticancerous properties (Morris, 1999). Similarly, 'canavanine' extracted from jackbean (Canavalia ensiformis) is also reported to be cytotoxic to human pancreatic cancer cells (Swaffer et al., 1995).

Sure and Read (1921) have detailed the biological analysis of seed of Georgia velvet bean (Stizolobium deeringianum). Ferris (1917) and Fain and Tabor (1921) have mentioned on the use of Mucuna as ruminant feed. Scott (1916) and Lamaster and Jones (1923) have reported use of Mucuna seeds as feed for dairy cows. Tweedie and Carew (1963) also reported the use of velvet beans as ruminant feed. Mucuna plant has been used in mixed cropping with maize and cowpea and the yield and chemical composition of fodder have been described by Singh and Relwani (1978). Harms et al. (1961) reported the influence of feeding various levels of velvet beans to chicks and laying hens. Species differentiation between Mucuna with reference to seedling morphology has been described by Sastraprajada et al. (1975). Mucuna pruriens has been extensively used as cover crop for enhancement of water infiltration, softening the soil, improvement of soil fertility and to suppress the weeds (Acanthospermum hispidum, Euphobia hirta, Senescio vulgaris, Oxygonum sinuatum, Schkuria pinnata, Richardia brasiliensis, Bidens pilosa, Sonchus oleraceae) (Osei-Bonsu et al., 1994; Mwangi et al., 2006)

3 MATERIALS AND METHODS

3.1. Description of the Study

The experiment was conducted in Bonga university research site of Kaffa Zone South Western Ethiopia during the Belg season (October-January) of 2022/23. Kaffa zone had abundant rainfall throughout the year; with mean annual rainfall ranging between 1600 to 2200mm and annual temperature vary from 16°C to 20°C. It has abundant rainfall distribution nearly throughout the months of the year. Main economic activity in the area is agriculture that contributes approximately 41% to the Gross Domestic Products. There are two main seasons in this study area, the wet season from March to November and the dry season from December to February (CSA, 2005).

3.2. Chemicals and Reagents

All chemicals and reagents were used for current study is analytical grade with high purity. The required chemicals and regents for conducting experimental analysis includes: Hydrochloric acid, ammonium hydroxide, phenolphthalein, glacial acetic acid, calcium chloride, sulfuric acid, sodium hydroxide, sodium chloride, potassium permanganate, sodium carbonate, N-benzoyl-DI-arginine, iron chloride, per chloric acid, molybdate, phosphomolbdate complex, methanol, Folin-ciocalteau reagents, ethanol, diethyl ether, n-butanol, DPPH, Distilled water and another types of chemicals and reagents was used for proximate analysis, mineral composition, phytochemical compounds and anti-nutritional factors from extracted *Mucuna prurience* seed.

3.4. Sample collection

3.4.1. Sample collection and pre-treatment

Velvet bean (*Mucuna prurience*) seed sample was collected from Bonga University project site and was packed in polyethylene plastic bags. Then the collected sample was transported to chemistry laboratory of Bonga University Ethiopia for further analysis. Before, starting sample preparation and analysis of the sample, carefully cleaning the collected *Mucuna pruriens* seed sample manually to remove all foreign matter, immature and damaged seed. Once, the seed is cleaned, washing the surface of the sample using tape water reputedly followed rinse with distilled water. Samples of collected matured and dry Mucuna prurience seed plants are described in figure 1 below.

3.5. Sample preparation

The dry and matured *Mucuna prurience* seed was carefully separated from the peel through manual and collected seed on dry and clean polyethylene plastic bags. Followed, washing the surface of seed samples using tap water and rinsed with distilled water. There are different types of food processing techniques for the removal of anti-nutritional factors and ready for consumptions purpose.

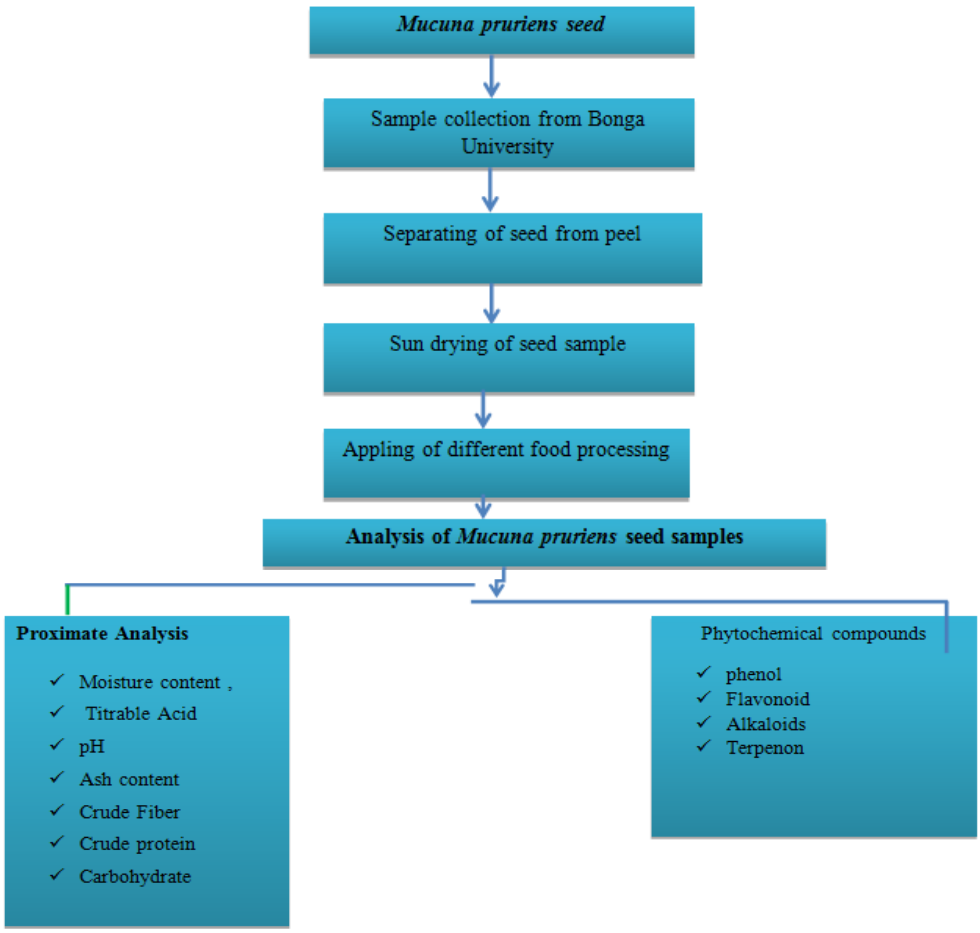


Figure 2: Experimental framework for analysis of *Mucuna prurience* seed sample.

Based on their familiarity in the community and literature report of food processing techniques for legume seed particularly *Mucuna prurience* three food processing (soaking, cooking and roasting) techniques was used for extraction of the samples and analysis of phytochemical compounds, mineral compositions, anti-nutritional factors and proximate analysis from the samples. At the end, over all the framework of sample preparation and analysis of the samples are described in **Figure 2** below.

3.5.1. Cooking of *Mucuna prurience* seed sample

According to[13] reported procedure with slightly modification, from whole collected dry, washed and clean *Mucuna prurience* seed samples (1.0 kg), weight 300.0 g of seed samples and boil with distilled water(1:10 W/V) for one hour at 60oC in hotplate. Then, filter the cooking sample and dry in an oven at 60oC for 24 hour. Then after, cool the sample on desiccator for an hour and grinding the dry sample using electrical miller. Followed, sieved the powder through 200 mm mesh and collected the powered sample on low density polyethylene plastic for further analysis.

3.5.2. Roasting of Mucuna prurience seed sample

Roasting techniques is one of the food processing techniques for the reducing of anti-nutritional factors and ready for consumption purpose of *Mucuna pruriens* seed. According to[13] reported procedure with sightline modification 200.0 g of the sample was weighted and transfer into metal pan. Then placed the sample contained metal pan into stove and roasting for 50 minutes. Then cool on desiccator for an hour and grinding using electrical miller. The powered was sieved through 200 mm mesh and collected the powered sample on low density polyethylene plastic for further analysis.

3.5.3. Soaking of Mucuna prurience seed sample

Similar to cooking and roasting/toasting, soaking is one of the food processing techniques used for reducing/eliminating/ of anti-nutritional factors from *Mucuna pruriens* seed. Therefore, from the whole collected and ready for sample preparations, 400.0 g of *Mucuna prurience* seed samples was soaked with distilled water (1:5, W/V) for three day at room temperature.[13] At 24 hour interval fresh water was replaced during the process. After three day the sample was filtered through Whatman filter paper No 42 and drying the sample for 24 hour at 60^{o}C in an oven. Finally, grinding the dry sample through electrical miller and collected on polyethylene plastic for further analysis.

3.6. Proximate analysis of *Mucuna prurience* seed sample

The proximate compositions (moisture, ash, crude protein, crude fibre, crude fat, dry matter and total carbohydrate) content from the extracted Mucuna pruriens seed sample was evaluated using Association of Analytical Chemists (AOAC) method.[18] According to the proximate analysis for each parameter are described below.

3.6.1. Determination of Moisture contents

From well-organized ground sample, weight 2.0 g of the sample transfer into constant and empty crucible. Then placed the sample contained crucible into drying oven and dry the sample for three hour at 105oC. Then after, remove the sample from the oven and cool in desiccator for an hour at room temperature and weighted the dry sample. Finally the moisture contents of the sample was calculated using below described equations.

$$\% \text{ Moisture content} = \frac{W2-W3}{W2-W2} * 100 \text{ ----------Eq(1)}$$

Where: W_1 = Mass of empty crucible (g)
W_2 = Mass of the sample before drying
W_3 = Mass of the sample after drying

3.6.2. Determination of total ash content

Weight 2.0 g of powder *Mucuna prurience* seed sample and transfer into constant weighted silica crucible dish. Then placed sample contained dish into muffle furnace and ignition of the sample at 550^{o}C for three hours until ash color was developed. Then removed dish from the furnace and cool in desiccator for hour in room temperature. Finally, weighted the residue (ash) and calculated the total ash contents from the sample that described equation 2 below.

$$\% \text{ ash} = \frac{W2-W3}{W1} * 100 \text{----------Eq(2)}$$

Where:
W_1 = Mass of empty crucible (g)
W_2 = Mass of the sample before drying
W_3 = weight of ash

3.6.3. Estimation of crude fiber

Crude fiber consists largely of cellulose & lignin (97%) with some mineral matters, 60-70% cellulose & 4-6% lignin.

During acid & subsequent alkali treatment, oxidative hydrolytic degradation of native cellulose & considerable degradation of lignin occurs. Residue obtain after final filtration is weighed, incinerated, cooled & weighed again. Loss in weight gives crude fiber contents.

1. Two g of sample was weighed & transferred in pre weighed crucible/beaker which was then placed on hot extraction unit/s & bath.
2. 150 ml of 1.25% H2SO4 is poured in to extractor from top (Acid wash).
3. Instrument was switched on & initial temp is set at 400˚C. Sample was allowed to boil for 40 min in acid. Then acid was drained by filtering suspension through a sintered filtration unit under vacuum. Residue on sintered disc was washed thrice with distilled water & dried.
4. Dried material was shifted to same beaker & 150 ml of 1.25% NaOH was poured in to extractor from top

(Alkali wash).

5. Instrument was switched on & initial temp is set at 400˚C. Sample was allowed to boil for 40 min in alkali. Then alkali was drained using sintered filtration unit as described above & samples were washed twice or thrice with distilled water. Residue was dried under vacuum.
6. Then residue was shifted to pre weighed crucible & residue was placed in hot air oven to get rid of any moisture.
7. Crucible were weighed & readings were recorded (CWBA= W1).

All crucibles were placed at 550˚C for ashing in a Muffle furnace. Crucible were cooled down after ashing & weighed (CWAA=W2) by fwg formula. Finally, weighted the ash and calculated the crude fiber from the sample using the following equation that described below.

% crude fiber = $\frac{W3}{W}$ **X100% ----------Eq(3)**

Where:
W= the sample weight
W_1 = initially weighted grounded sample
Crucible Weight After Ashing (CWAA)= W2
Change in weight W3= (W1-W2)

3.6.5: Determination of crude protein from *Mucuna pruriens* seed sample

The analysis of protein content was examined from the extracted Mucuna pruriens seed sample using the micro kjeldahl method according to the official method 979.09 of the AOAC (2000). Then to analysis of protein content from the sample three steps was applied: Digestion, Distillation and Titration. Each step is briefly described below under here. 0.5 g of *Mucuna pruriens* seed sample was weighted in a clean tecator tube and placed in tractor rack. In this sample, 5 mL of concentrated sulfuric acid was added carefully for digestion of the samples. Followed, 2.5 mL of hydrogen peroxide was added in a step wise manner to each sample tube. Then shake the sample contained flask tube shake carefully for a minute by hand and put it back to the rack. In this mixture of sample add 2.5 g of catalyst of copper sulfate and potassium sulfate. Then the sample contained flask was let to stand for 20 minute before digestion.

A. Digestion process

The sample tubes was placed in a digester after the working temperature has reached at 370^0C and the digestion process has continued until clear solution was observed. The sample tubes will take out, placed in the rack and allowed to cool in fume hood. Later on, 50ml of distilled water was added into the sample tubes in order to avoid precipitation of sulfate.

B. Distillation process

Under this step, 20ml of 35% NaOH was added to neutralize sulfuric acid and this enables for the release of ammonia. A 250ml Erlenmeyer flask containing 25 ml of 4% H_3BO_3, 30 ml of distilled water and 3 drops of methyl red indicator solution was placed as receiver on the distillation unit. The distillation process was continued until the volume of the distillate reached between 200ml and 250ml.

C. Titration process

Similar to the above described two steps, under this step, the distillate was finally titrated with standardized 0.1N of HCI until the appearance of the first pink color. At this point the amount of consumed HCI was immediately recorded. Likewise, the blank reagent was run to subtract the reagent Nitrogen from the sample Nitrogen. Finally, the amount of protein content from the extracted sample was calculated using equation 4 below

Crude protein (%): $\frac{(V2-V1)*N*14.01*6.25}{10\ W}$----------Eq (4)

Where
V_1= Volume (mL) of HCl required for the blank reagents
V_2 = Volume (mL) of HCl solution required for sample test
N= Normality of HCL
W= Weight of the sample

D) Crude fat

The method is based on the principle of lipid solubility in non-polar solvents. Fat is extracted from the seed sample with an organic solvent, typically petroleum ether or hexane. The solvent is then evaporated, and the extracted fat is dried and weighed. The weight of the extracted fat is expressed as a percentage of the original sample weight (AOAC, 2005).

Calculate the weight of the fat by subtracting the weight of the empty flask from the weight of the flask plus fat. Calculate the percentage of crude fat in the original sample using the following formula:
Crude Fat (%) = (Weight of fat / Weight of sample) × 100

3.7. Determination of total carbohydrate

The total carbohydrate content of Mucuna purines seed sample was calculated by using of the described equation below (**Equation 7**)
Total carbohydrate = 100 – [% moisture + % ash + %fiber + % protein + % fat] --- Eq (7)

3.9. Qualitative tests for preliminary phytochemical screening

3.9.1. Extraction of *Mucuna purines* seed samples for phytochemical compound analysis

The equimolar mixture ethyl ether and n-hexane extract of ***Mucuna purines seed*** was subjected to preliminary phytochemical screening of various plant constituents (Harborne, 1998; Kokate, 2001).

A) Test for Alkaloids

(i) Dragendroff's test

Dragendroff's reagent: Eight grams of $Bi(NO_3)_3$ $5H_2O$ was dissolved in 20 ml of HNO_3 and 2.72 g of potassium iodide in 50 ml of H_2O. These were mixed and allowed to stand until KNO_3 crystals formed. The supernatant was decanted off and made up to 100 ml with distilled water.

Procedure: To 0.5 ml of the extract was added to 2 ml of HCl. To this acidic medium, 1 ml of Dragendroff's reagent was added. An orange or red precipitate produced immediately indicates the presence of alkaloids.

(ii) Wagner's test

Wagner's reagent: 1.2 g of iodide and 2.0 g of potassium iodide were dissolved in 5 ml of Sulphuric acid and the solution was diluted to 100 ml.

Procedure: 10ml of the extract was acidified by adding 1.5% v/v of HCl and a few drops of Wagner's reagent.

Formation of yellow or brown precipitate confirmed the presence of alkaloid.

(iii) Mayer's test

Mayer's reagent: 1.36 g of mercuric chloride was dissolved in 60 ml of distilled water and 5 g of potassium iodide in 10 ml of water. The two solutions were mixed and diluted to 100 ml with distilled water.

Procedure: 1.2 ml of the extract was taken in a test tube, 0.2 ml of dilute hydrochloric acid and 0.1 ml of Mayer's reagent were added. Formation of yellowish buff coloured precipitate confirmed the presence of alkaloid.

B) Test for Flavonoids

(i) Shinoda's test: In a test tube containing 0.5 ml of extract, 5-10 drops of diluted HCl and small piece of ZnCl or magnesium were added and the solution was boiled for a few min. In the presence of flavonoids, reddish pink color was produced.

(ii) Alkaline Reagent Test: To 1.0 ml of the extract, a few drops of dilute sodium hydroxide were added. An intense yellow color was produced in the plant extract, which become colourless on addition of a few drops of dilute acid indicates the presence of flavonoids.

C) Test for Carbohydrates

A small quantity of the extract was dissolved separately in 4 ml of distilled water and filtered. The filtrate was subjected to Molisch's test to detect the presence of carbohydrates.

Molisch's test: Filtrate was treated with 2-3 drops of 1% alcoholic a-naphthol solution and 2 ml of Conc. H_2SO_4 was added along the sides of the test tube. Appearance of brown ring at the junction of two liquids shows the presence of carbohydrates.

D) Test for Glycosides

The extract was hydrolysed with HCl for few hours on a water bath and the hydrolysate was subjected to Legal's or Borntrager's test to detect the presence of glycosides.

(i) Legal's test: To the hydrolysate add 1 ml of pyridine and a few drops of sodium nitroprusside solutions were added and then it was made alkaline with sodium hydroxide solution. Appearance of pink to red colour shows the presence of glycosides.

(ii) Borntrager's test: Hydrolysate was treated with chloroform and then the chloroform layer was separated. To this equal quantity of dilute ammonia solution was added. Ammonia layer acquires pink color, shows the presence of glycosides.

E) Test for Saponins

(i) The extract was diluted with 20 ml of distilled water and it was agitated in a graduated cylinder for 15 min. The formation of 1 cm layer of foam shows the presence of saponins.

(ii) 1 ml of the extract was treated with 1% lead acetate solution. Formation of white precipitate indicates the presence of saponins.

F) Test for Tannins

(i) Ferric chloride test: To 1-2 ml of the extract and a few drops of 5% aqueous $FeCl_3$ solution was added. A violet colour formation indicates the presence of tannins.

(ii) Lead acetate test: In a test tube containing about 5.0 ml of the extract and a few drops of 1% lead acetate was added. A yellow precipitate was formed, indicates the presence of tannins.

(iii) 5 ml of the extract was treated with 1 ml of 10% aqueous potassium dichromate solution. Formation of yellowish brown precipitate indicated the presence of tannins.

G) Test for Phytosterol

The extract was refluxed with solution of alcoholic potassium hydroxide till complete saponification takes place. The mixture was diluted and extracted with ether. The ether layer was evaporated and the residue was tested for the presence of phytosterol.

(i) Libermann Burchard test: The residue was dissolved in few drops of diluted acetic acid, 3 ml of acetic anhydride was added followed by a few drops of Conc. H_2SO_4. Appearance of bluish green colour shows the presence of phytosterol.

(ii) Salkowski test: 10 mg of the extract was dissolved in 1 ml of chloroform, 1 ml of Conc. H_2SO_4 was added carefully along the sides of the test tube. The red colour was produced, indicating the presence of sterols.

H) Test for Triterpenoids

(i) Libermann Burchard test: 10 mg of the extract was dissolved in 1 ml of chloroform, 1 ml of acetic anhydride was added following the addition of 2 ml of Conc. H_2SO_4. Formation of reddish violet colour indicates the presence of triterpenoids.

(ii) Noller test: 5 mg of the extract was dissolved in 2 ml of 0.01% anhydrous stannic chloride in pure thionyl chloride. A purple colour formed then changed to deep red after few minutes, indicates the presence of triterpenoids.

I) Test for Proteins and Amino Acids

(i) Ninhydrin test: 1.0 ml of the extract was treated with few drops of Ninhydrin reagent (Triketo hydrindene hydrate). Appearance of purple colour shows the presence of amino acids.

(ii) Biuret test: Equal volumes of 5% NaOH solution and 1% copper sulphate solution were added to 1.0 ml of the extract. Appearance of purple color shows the presence of proteins.

J) Test for Anthraquinones

5 ml of the extract solution was hydrolyzed with dil. H_2SO_4 and extracted with benzene. 1 ml of dilute ammonia was added to it. Rose pink coloration indicated the positive response for anthraquinones.

Estimation of Nitrogen percentage

Nitrogen is a major nutrient required by plants & is essential for cell division, expansion & growth. Sample is digested by boiling with concd H_2SO_4 in presence of catalyst $CuSO_4$. Digestion converts all the N to NH_3 which is trapped as $(NH_4)_2SO_4$. Completion of digestion stage is generally recognized by formation of clear solution. NH_3 is released by addition of excess NaOH is removed by steam distillation. It is collected in boric acid & titrated with standard HCl using methylene blue as an indicator.

1. 50 mg of dried plant powder was taken in Kjeldahl digestion tube (10 ml). 50 mg of catalyst $CuSO_4$ with 2.5 ml of concd H_2SO_4 was added to tube.
2. Digestion tubes were then placed in s& bath digestion unit at 400˚C temp ensuring tubes are placed straight. Digestion was continued till brown or black colour of sample disappears & clear solution is formed for 6-8 hs for completion. Sample was left for cooling at room temp.
3. Steam was generated into Kjeldahl distillation unit (ASGI, India). Once steam is generated at good pace, digested material was poured into distillation tube followed by addition of 10 ml of 40% NaOH slowly. During distillation NH_3 was released due to addition of NaOH.
4. Condenser outlet of distillation unit was dipped in flask which contains 10ml of 4% boric acid & few drops of methyl red indicator. NH_3 is trapped by boric acid & due to change in pH, solution turns colorless. Thus distillation was completed.
5. Boric acid with trapped NH_3 was titrated against 0.1 N HCl. Boric acid blank was also run & the titration was carried out like that of the sample. N contents in plant sample were calculated using following formula.

$$N\,(\%) = \frac{(S - B) \times \text{Normality of HCl} \times 100}{1.4007\ W}$$

Where: - S= Reading of sample (amount of HCl required for sample).
B= Blank reading of sample (amount of HCl required for blank).
W= Weight of sample in gr used for digestion

3.10. Test of oxalate from *Mucuna prurience* seed

Oxalate content of the Mucuna prurience seed sample was determined using the method of[44] with slight modification. To determine the oxalate content from the sample the following steps was used during the conducting of experiments (digestion, precipitation and permanganate titration). Briefly, in digestion steps, two gram of powder sample was suspended in 200 ml of distilled water contained in a 250-ml volumetric flask; 10 ml of 6M HCl was added and the suspension digested at 80°C for 2 hour, followed by cooling, and then made up to 250 ml before filtration. Oxalate precipitation: Duplicate portions of 125 ml of the filtrate was measured into a beaker and four drops of methyl red indicator added, followed by the addition of concentrated NH_4OH solution (drop wise) until the test solution changed from its salmon pink color to a faint yellow color.

3.11 The allelophatic effect: The allelophatic effect **of** *mucuna prurience* on haricot bean and coffee was evaluated by observing the growth performance of both crops.

3.12 Mineral Exploitation; The Mineral Exploitation of mucuna prurience was evaluated y soil testing before sowing and after harvesting.

3.13. Statistical analysis

All analyses were done in triplicate. One-way analysis of variance (ANOVA) was used to test the effect of the growing region on the mean concentrations of the different chemical constituents determined. Data analysis was carried out using the statistical software package SPSS 20 (IBM Corporation, USA). Differences was considered significant when $\alpha<0.05$.

4. RESULTS AND DISCUSSIONS

The qualitative phytochemical analysis of the Raw, Boiled, Rosted and Socked plant extract revealed the presence of several bioactive compounds. The following phytochemicals were detected: alkaloids, carbohydrates, reducing sugars, glycosides, proteins and amino acids, flavonoids, phenolic compounds, tannins, phlobatannins, saponins, terpenoids, triterpenoids, anthocyanins, carboxylic acids, and resins. Phytosterols, cholesterol, and anthraquinones were not detected. The results of the phytochemical screening are summarized in Table 1.

Table 1: Effect of Processing on Phytochemical Composition of Mucuna pruriens Seeds.

Test	Raw	Rosted 1hr 100^0C	Boild for 2hr at 100^0C	Soaked for 3 days
Alkaloids	++	+	+	+
Carbohydrates	++	+	+	+
Reducing Sugars	++	+	+	+
Glycosides	+	+	+	+
Proteins and Amino Acids	++	+	+	+
Flavonoids	++	+	+	+
Phenolic Compounds	++	+	+	+
Tannins	+++	+	+	+
Phlobatannins	+	+	+	+
Saponins	++	+	+	+
Terpenoids	+	+	+	+
Triterpenoids	+	+	+	+
Anthraquinones	-	+	+	+
Anthocyanins	+	+	+	+
Carboxylic Acids	+	+	+	+
Resin	+	+	+	+

KEY: + Low presence. ++ Moderately presence. +++ High presence

DISCUSSION

Mucuna prurience also known as velvet bean, is a tropical legume that has been used in traditional medicine for centuries (Katzenschlager R., *et al*, 2004). As indicated in the different (Ezegbe C.C., *et al*, 2023) studies, the seeds of Mucuna prurience are rich in bioactive compounds, including alkaloids, carbohydrates, reducing sugars, glycosides, cardiac glycosides, proteins and amino acids, flavonoids, phenolic compounds, tannins, phlobatannins, saponins, phytosterols, cholesterol, terpenoids, triterpenoids, quinones, anthraquinones, anthocyanins, leuconthocyanins, carboxylic acids, emodin, and resins (Mayuri Singh, et al, 2022). This study also interested to determine the presence of some essential phytochemicals in muccuna seed in the Ethiopian agro ecology.

The results current study on the phytochemical screening of Mucuna pruriens seed extract reveal the presence of various important compounds. The detection of carbohydrates, including monosaccharaides and reducing sugars, is indicative of the seed's potential nutritional value (Aneke V.I et al, 2019). The presence of glycosides is also significant, as these compounds have been associated with various biological activities, including antioxidant and antimicrobial properties. The findings are consistent with the aim of the study, which was to scrutinize the presence of nutritional and phytochemical contents of the Mucuna pruriens seed in the study area.

The results of this study indicate the detection of tannins, phlobatannins, saponins, phytosterols, and terpenoids in the seed extract. The presence of tannins and phlobatannins in the seed extract is in line with previous studies (Govindan Sh., 2023) that have identified these compounds in Mucuna prurience seeds.

The results indicate a clear impact of processing on the phytochemical profile. Roasting, boiling, and soaking generally reduced the presence of most phytochemicals compared to the raw sample. For example, the intensity of tannins decreased from high (+++) in the raw sample to low (+) in all processed samples. This reduction could be attributed to the degradation or leaching of these compounds during processing.

The presence of terpenoids in the seed extract is also consistent with existing research (Ashidi, J.S, 2022), emphasizing the potential health-promoting properties of these compounds. According to the study conducted by (Bailey L.H, 2014), Terpenoids have been linked to various pharmacological activities, including anti-inflammatory and anticancer properties. Finally the results of this study indicate the detection of terpenoids, anthocyanins, carboxylic acids and resins in the seed extract. These findings contribute to a deeper understanding of the seed's phytochemical composition and its potential applications in the food and pharmaceutical industries.

The result of the phytochemical screening shown in the above table 4.1., the absence of Phytosterols, Cholesterols, and Anthraquinones in muccuna pruriens extracts along with method used. Contrarily, the study conducted by (Krishavan M. 2017), revealed that muccuna pruriens seed contain steroids. Another study conducted by (Minari J.B. et al 2015), also indicates the presence of Anthraquinones and Steroids in Mucuna prurience seed. Additionally, the study conducted by (Ashidi J.S., et al, 2022), confirmed that the presence of Steroids in muccuna seed. Therefore to conclude the absence of Anthraquinones, Steroids and Triterpenes in the Mucuna seed of study area, it needs further analysis followed by changing screening techniques and sophisticated instruments. Nevertheless, the result of the

current study indicates the absence of these phytochemicals. This result was supported by the study report of (Kumar A. et al, 2009) Steroids and saponins were not detected in methanolic extract of Mucuna seeds.

Alkaloids: These are a group of naturally occurring chemical compounds containing mostly basic nitrogen atoms. The presence of alkaloids in Mucuna pruriens seeds has been reported in several studies (Ashidi J.S., et al, 2022 and Ezegbe C.C., et al, 2023) Alkaloids are known for their diverse biological activities, including antimicrobial, anti-inflammatory, and anticancer properties (Krishnaveni M et al 2017). They have diverse pharmacological effects (Krishnaveni & Hariharan, 2017). The detection of alkaloids in the seed extract further emphasizes the potential health-promoting properties of Mucuna prurience seeds. This study revealed that in Mucuna pruriens, the intensity of alkaloids decreased with all processing methods (roasting, boiling, and soaking).

Carbohydrates, Glycosides and Reducing Sugars: The presence of carbohydrates Glycosides and reducing sugars in Mucuna prurience seeds has been reported in studies of (Lampariello L. R., et al, 2011). These compounds are known for their potential nutritional and medicinal significance, and their presence in the seed extract underscores its potential health benefits. These are essential biomolecules that serve as a primary source of energy. The table indicates that the intensity of carbohydrates decreased upon processing (Ferdous et al., 2021).

Proteins and **Amino Acids**: The presence of proteins and amino acids in Mucuna prurience seeds has been reported in several studies (Minari J.B., et al, 2015). These are the building blocks of proteins, essential for various biological functions. Proteins and amino acids are essential for human nutrition and are the building blocks of tissues and muscles. The present study revealed that the processing led to a decrease in their intensity.

Flavonoids: These are polyphenolic compounds with antioxidant properties. The presence of flavonoids in Mucuna prurience seeds has been reported in the studies of (Ashidi, J. S, et al 2022). Contrarily, the study report of (Mayuri Singh, et al 2022), showed that Mucuna prurience seed do not contain Flavonoids. Flavonoids are known for their diverse biological activities, including antioxidant, anti-inflammatory, and anticancer properties (Ashidi, J.S, 2022). According to study of (Shanmugavel G and Krishnamoorthy G, 2018), revealed that Mucuna pruriens seed as rich source of nutrients and phytochemicals and the presence of alkaloids, flavonoids, glycosides, saponins, steroids, tannin and terpenoids (Nwaoguikpe R.N., et al 2011). Then the current study also confirmed that presence of flavonoids in muccuna pruins seed and processing reduced their intensity (Ferdous et al., 2021).

Phenolic Compounds: These are a large class of aromatic compounds with antioxidant and other beneficial properties. The presence of phenolic compounds in Mucuna pruriens seeds has been reported in the studies of (Ashidi, J. S, et al, 2022). Phenolic compounds are known for their diverse biological activities, including antioxidant, anti-inflammatory, and anticancer properties (Minari J.B, 2015.). The detection of phenolic compounds in the seed extract further emphasizes the potential health-promoting properties of Mucuna pruriens seeds. According to the study conducted by (Ezegbe C.C., et al, 2023), raw Mucuna pruriens seed contains phenol. Another study conducted by (Makoye P.M., et al, 2020), also revealed that the presence of phenolic compounds in muccuna purine seed extracts. The current study confirms that the presence of flavonoids and processing decreased their intensity (Ferdous et al., 2021).

Tannins: These are polyphenols known for their astringent properties and anti-nutritional effects. The presence of tannins in Mucuna pruriens seeds has been reported in several studies (Murthy S. N., et al 2016 and Kumar A., et al, 2009). Tannins are known for their potential health benefits, including antioxidant, anti-inflammatory, and antimicrobial properties (Ezegbe C.C., et al, 2023). A significant reduction in tannin intensity was observed after all processing methods, which is a desirable outcome.

Phlobatannins: Phlobatannins are a type of condensed tannin, which are complex polyphenolic compounds derived from the polymerization of flavanols. They are known to contribute to the astringency and color of certain plants (Nata et al., 2022; Theansungnoen et al., 2021). While tannins, in general, have anti-nutritional properties, some studies suggest that phlobatannins may also possess antioxidant and other biological activities. In this study, the presence of phlobatannins remained consistent across raw and processed samples. This suggests that the processing methods used did not significantly alter the levels of these compounds. Further research may be needed to fully elucidate the specific role and potential benefits or drawbacks of phlobatannins in Mucuna pruriens.

Saponins: These are glycosides with soap-like properties. The presence of saponins in Mucuna pruriens seeds has been reported the study report of (Kumar A. et al, 2009). Obstinately, the report of studies (Mayuri Singh, et al 2022), indicated the absence of saponins in muccuna seed. The present study confirmed the presence of saponins in raw muccuna seed extract. Saponins are known for their diverse biological activities, including antioxidant, anti-inflammatory, and anticancer properties (Tavares R.L., et al, 2015). According to the result of this study Processing reduced their intensity (Krishnaveni & Hariharan, 2017).

Terpenoids: Terpenoids are a large and diverse class of organic compounds, produced primarily by plants. They are derived from isopentenyl pyrophosphate (IPP) or its isomer, dimethylallyl pyrophosphate (DMAPP). Terpenoids play various roles in plant metabolism, including defense against herbivores and pathogens, and as signaling molecules. They are also known for their medicinal properties, including anti-inflammatory, antioxidant, and anticancer activities (Dudareva et al., 2013; Gershenzon & Dudareva, 2007). The presence of terpenoids remained consistent across raw and processed samples.

Triterpenoids: Triterpenoids are a subclass of terpenoids, consisting of six isoprene units. They are found in various plants and have diverse structures and functions. Like other terpenoids, triterpenoids have been shown to possess a range of pharmacological activities, including anti-inflammatory, antimicrobial, and cytotoxic effects (Cichewicz & Kouzi, 2003; Mahato et al., 1994). The consistent presence of triterpenoids in both raw and processed samples suggests that these compounds are relatively stable under the processing conditions used in this study.

Anthraquinones: Anthraquinones are a class of organic compounds, many of which are naturally occurring pigments with a wide range of colors, from red and orange to yellow. In plants, they often serve as protective agents, deterring herbivores and pathogens. Anthraquinones are known to have various medicinal properties, including laxative, antimicrobial, and anti-inflammatory effects (Bruneton, 1999; Thomson, 1987). The observation that anthraquinones were absent in the raw seeds but appeared after processing suggests that these compounds may be formed or released from a bound form during the roasting, boiling, and soaking processes. This could be due to the breakdown of complex molecules or the release of anthraquinones from the plant matrix (Igbinaduwa & Anoh, 2012).

Anthocyanins: Anthocyanins are a group of water-soluble pigments that belong to the flavonoid family. They are responsible for the red, purple, and blue colors found in many fruits, vegetables, and flowers (Gould et al., 2009). Beyond their role as natural colorants, anthocyanins are potent antioxidants, capable of scavenging free radicals and protecting cells against oxidative stress. Research suggests that anthocyanins may have a range of health benefits, including reducing the risk of cardiovascular disease, improving cognitive function, and exerting anti-inflammatory effects (Rahman et al., 2006; Tsuda, 2012). The consistent presence of anthocyanins in both raw and processed samples indicates that these pigments are relatively stable under the processing conditions used in this study.

Cardiac Glycosides: The presence of cardiac glycosides in Mucuna pruriens seeds has been reported in several studies (Mattioli R. 2020 and Tavares R. L., et al. 2020). Additionally, the study report of (minari J.B., et al 2015), also revealed the presence of Cardiac glycosides in muccuna seed. Cardiac glycosides are known for their potential medicinal properties, including their use in the treatment of heart failure and arrhythmias (Ezegbe C.C., et al, 2023). The result of present qualitative analysis of raw muccuna seed confirmed the presence of Cardiac Glycosides in muccuna seed. The detection of cardiac glycosides in the seed extract further emphasizes its potential medicinal significance.

Resins: Resins are complex mixtures of solid or semi-solid amorphous organic compounds. In plants, resins often serve protective functions, such as sealing wounds and deterring herbivores and pathogens due to their sticky or toxic properties (Langenheim, 2003). The presence of resins in Mucuna pruriens seeds has been reported in the studies of (Mayuri Singh, et al 2022). The composition of resins can vary significantly between plant species, but they commonly include terpenoids, phenolic compounds, and volatile oils (Gershenzon & Dudareva, 2007). Resins have been used traditionally for various purposes, including medicinal applications, incense, and varnishes (Langenheim, 2003). The consistent presence of resins in Mucuna pruriens across different processing methods suggests that these compounds are stable and not significantly affected by heating or soaking.

The presence of these bioactive compounds in the seed extract further emphasizes the potential health benefits of Mucuna pruriens seeds. In conclusion, the phytochemical screening results, supported by existing literature, provide robust evidence of the presence of various bioactive compounds in Mucuna pruriens seed extract. These findings underscore the significance of Mucuna pruriens seeds as a valuable source of natural products with potential medicinal and nutritional applications. These compounds have been associated with a wide range of biological activities, including antioxidant, antimicrobial, and anti-inflammatory properties.

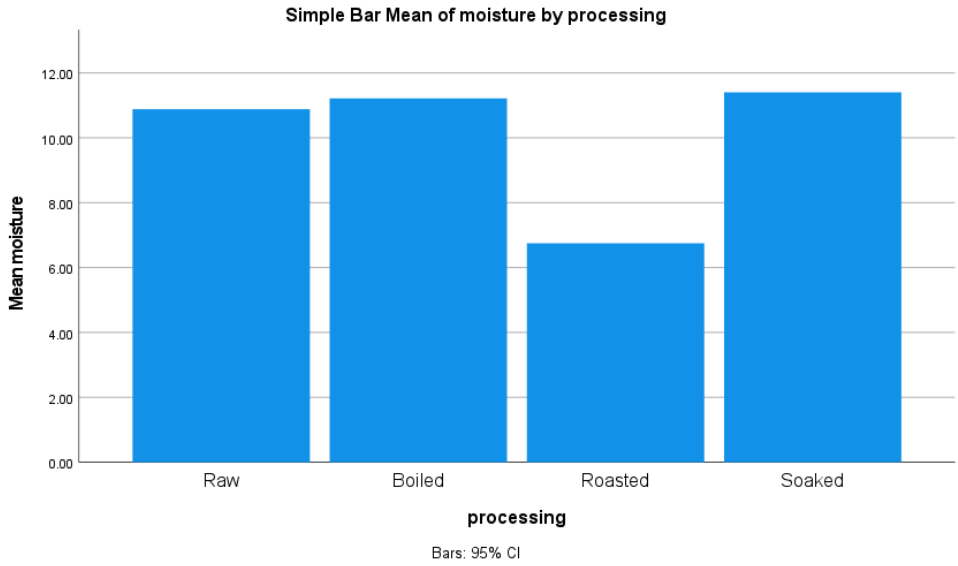
Table 2: Effect of Processing Methods on the Nutritional and Physicochemical Properties of the Mucuna Prunes seed.

Parameters	processing	Mean	Minimum	Maximum
Moisture	Raw	10.88±1.28	9.83	12.32
	Boiled	11.21±.87	10.24	11.93
	Roasted	6.75±.54	6.21	7.29
	Soaked	11.4±.48	10.97	11.92
Protein	Raw	28.22±.23	27.98	28.45
	Boiled	22.21±1.18	21.05	23.42
	Roasted	24.89±.44	24.57	25.40
	Soaked	21.97±.56	21.37	22.50
Crude fiber	Raw	3.68±.15	3.57	3.86
	Boiled	3.60±.29	3.27	3.83
	Roasted	3.80±.52	3.26	4.30
	Soaked	3.79±.17	3.61	3.96
Crude fate	Raw	3.52±.69	2.96	4.30
	Boiled	2.51±.08	2.43	2.60
	Roasted	4.75±.23	4.48	4.91
	Soaked	2.37±.21	2.12	2.51
Carbohydrate	Raw	59.92±.64	59.23	60.50
	Boiled	67.33±.77	66.46	67.92
	Roasted	57.60±5.5	53.94	64.00
	Soaked	67.71±.74	67.08	68.53
PH	Raw	6.443	6.4	6.5
	Boiled	6.397	6.4	6.4
	Roasted	6.417	6.4	6.5
	Soaked	6.427	6.4	6.5
Ash	Raw	4.5600	3.91	5.26
	Boiled	3.7100	2.98	4.24
	Roasted	3.3967	2.93	4.24
	Soaked	3.1600	2.98	3.40

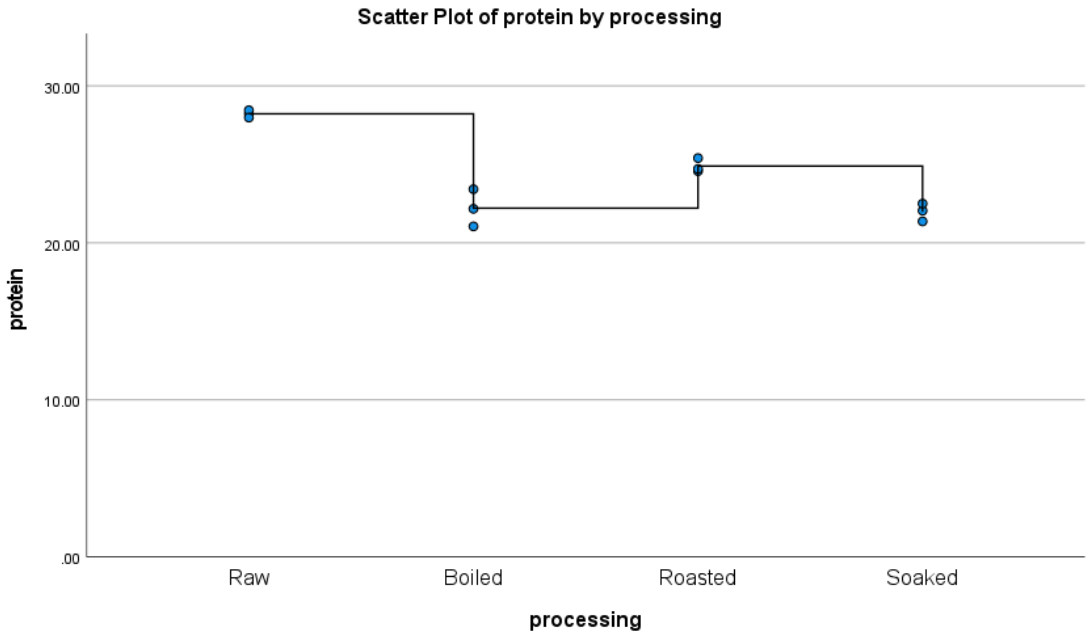
4. Physicochemical parameter

The results of this study clearly demonstrate the significant impact of boiling, roasting, and soaking on the nutritional and physicochemical composition of the Mucuna purines seed. According to the studies conducted by (Ezgbe C.C. *et al*, 2023), the proximate composition of the raw M. prurience seed contained 10.99% moisture, 3.82% ash, 25.34% crude protein, 4.69% crude fiber. The report of the other scholar (Govindan Sh., 2018), revealed that the proximate composition of the raw M. prurience seed contained moisture with 9.8%, protein with 12.1%, 28.2% proteins. The current study revealed the range of physicochemical composition of Mucuna prunes seed under different processing methods as shown table ---- above.

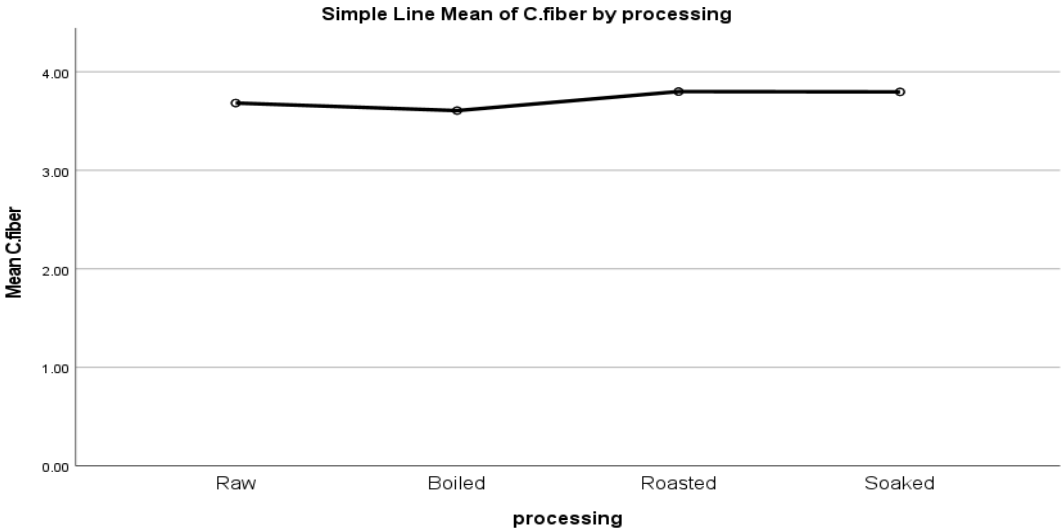
Moisture Content: Raw samples exhibited a moisture content ranging from 9.83% to 12.32% (mean 10.88%). The moisture content of the seed was found to range from 10.99% to 18-30% according to studies of (Ezegbe C. C. et al, 2023 and Alonge, A.F. et al. 2017). Additionally, Mucuna Pruriens Seed is reported to have moisture content below 9.8 ± 0.04 in the study of Tavares L. R. et al 2015). Roasting led to a substantial reduction in this range (6.21-7.29%, mean 6.75%), consistent with the dehydrating effect of dry heat. Similar reductions have been reported in studies on roasted grains and nuts (Araghi *et al*., 2011). Conversely, boiling resulted in a slight increase in the moisture range (10.24-11.93%, mean 11.21%), and soaking showed a similar trend (10.97-11.92%, mean 11.40%). This increase due to water absorption during boiling and soaking aligns with findings in legumes and cereals (Fellows, 2017)



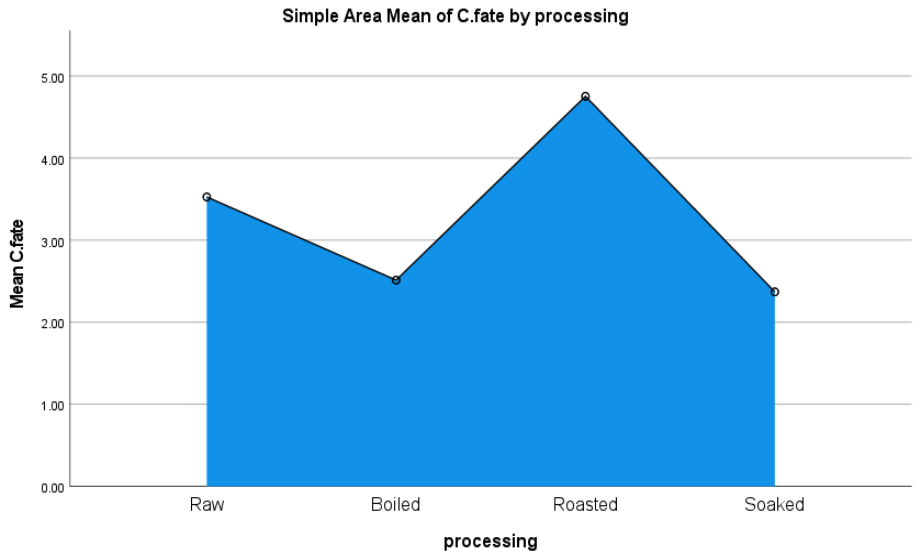
Protein Content: The protein composition of the seed indicates that it contains approximately 25.34% to 43.12% crude protein according to the study report of (Ezegbe C. C. et al 2023 and Tavares R.L. et al 2015). Additionally, a study on the chemical composition of the seed mentions a crude protein content of 314.4 g/kg, which is equivalent to 31.44%. (Perumal Siddhuraju 1996). In this study, the raw food matrix had a protein content ranging from 27.98% to 28.45% (mean 28.22%). Boiling and soaking both led to a decrease in this range (boiled: 21.05-23.42%, mean 22.21%; soaked: 21.37-22.50%, mean 21.97%). This reduction is likely due to the leaching of soluble proteins into the aqueous medium, a phenomenon observed in studies on boiled and soaked legumes where protein loss into the cooking water was significant (Singh & Jambunathan, 1981). Both boiling (22.21%) and soaking (21.97%) caused a considerable reduction in protein content compared to the raw sample (28.22%) (e.g., Abu-Ghannam & McKenna, 1997; Singh et al., 1987). The decrease was more pronounced with boiling and soaking than with roasting (24.89%) (e.g., Mbithe et al., 2002). This suggests that water-based processing methods are more likely to lead to protein loss, possibly through leaching (Cheftel et al., 1989), compared to dry heat methods like roasting, where protein denaturation might be the primary alteration (Fennema, 1996).



Crude Fiber: The crude fiber content in Mucuna prurience seed reported by the study of (Ezegbe C. C. et al 2023) was approximately 4.69%. This report was supported by the findings of a study which reported the crude fiber content of the seed as 4.69%. Another source study conducted by (Govindan Sh., 2023), also confirms a similar crude fiber content of 13.3%. Therefore, it can be concluded that the crude fiber content in Mucuna prurience seed is approximately range from 4.69% - 13.3 ± 0.18%. in this study the crude fiber content remained relatively stable across all treatments, with the raw samples ranging from 3.57% to 3.86% (mean 3.68%) and processed samples showing similar ranges (boiled: 3.27-3.83%, mean 3.60%; roasted: 3.26-4.30%, mean 3.80%; soaked: 3.61-3.96%, mean 3.79%). This stability suggests that the structural components of fiber are not significantly affected by these common processing methods, consistent with some research on fiber stability during cooking ((Elleuch et al., 2011).

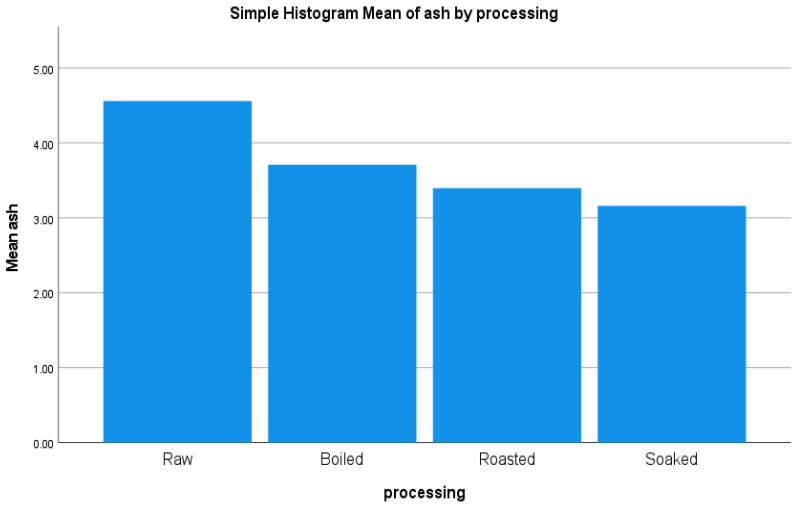


Crude Fat: The raw food matrix exhibited a crude fat content ranging from 2.96% to 4.30% (mean 3.52%). Roasting (4.75%) led to a notable increase in crude fat content compared to the raw sample (3.52%) (Lāina o Ghōṣha, 2012). Conversely, boiling (2.51%) and soaking (2.37%) resulted in a significant decrease in crude fat (e.g., Kolapo & Sanni, 2005; Reddy et al., 1986). This suggests that roasting can concentrate fat due to moisture loss (Potter & Hotchkiss, 1995), while water-based methods might lead to some fat loss, possibly through rendering or leaching (Kinsella, 1979). The effects of boiling and soaking on fat content were quite similar and opposite to that of roasting (Yīngxiǎng o Chéngfèn 2008).



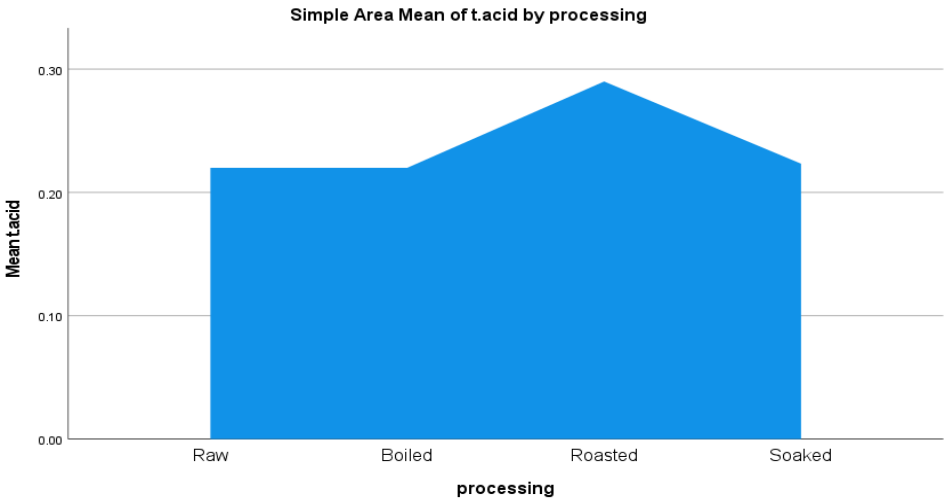
Carbohydrate Content: Raw samples had a carbohydrate content ranging from 59.23% to 60.50% (mean 59.92%). Boiling (range: 66.46-67.92%, mean 67.33%) and soaking (range: 67.08-68.53%, mean 67.71%) both showed a significant increase in the carbohydrate range, likely a relative increase due to the loss of protein, fat, and some minor components, coupled with slight moisture absorption. Boiling (67.33%) and soaking (67.71%) resulted in a substantial increase in carbohydrate content compared to the raw sample (59.92%) (Sōbahāna o Choudhurī, 2001). This increase is likely a relative effect due to the loss of other components like protein and fat, coupled with some water absorption (Shimkevich, 2007). Roasting (57.60%) showed a slight decrease in carbohydrate content (e.g., Ibanoglu, 2002). Thus, boiling and soaking appear to enhance the proportion of carbohydrates (Agrabāla o Gupta, 2003), while roasting might lead to some carbohydrate degradation or loss (Van Boekel, 2006).

pH: The pH values remained relatively stable across all treatments, with a narrow range observed in raw (6.4-6.5, mean 6.443), boiled (6.4-6.4, mean 6.397), roasted (6.4-6.5, mean 6.417), and soaked (6.4-6.5, mean 6.427%) samples. This consistency suggests that these processing methods, under the conditions of this study, do not significantly alter the overall acidity or basicity of the food matrix. Similar stability in pH has been reported in some cooked vegetables (e.g., Barbosa-Cánovas & Tel'nym, 2005), although pH changes can be more pronounced in fermentation or more extreme heat treatments.



Ash Content: Boiling (3.71%), roasting (3.40%), and soaking (3.16%) all led to a reduction in ash content compared to the raw sample (4.56%) (Goryachev o Petrov, 2009; Okaka & Okaka, 2001; Osman, 1991). Soaking resulted in the most significant decrease in ash content (e.g., Davies, 1987), followed by roasting and then boiling. This suggests that soaking, involving prolonged contact with water, might leach out more minerals than boiling (shorter water contact) or roasting (dry heat) (Reddy et al., 1989). The reduction in ash content during roasting could be due to the volatilization of some minerals at high temperatures or the formation of insoluble complexes. The wider range in raw and roasted samples might reflect natural variability in mineral content.

Titrable Acid:- According to a study, the titrable acidity of Mucuna prurience (velvet bean) seed was determined by titrating 20 ml of distilled water against 0.1M NaOH using phenolphthalein as an indicator (Hariom S., et al, 2022). The titre value was then used to calculate the titratable acidity as a percentage. This study shows that the Titrable acid content of raw Mucuna prurience seed is 0.25±.01%. According to the report of study conducted by (Ifesan, B O T. et al, 2017), indicated the total titratable acidity content of Mucuna is (7.01-22.72%). Another study conducted by (Yannick D.M. et al, 2016), report shows that the Titrable acid of Mucuna prurience seed ranges from 0.23 – 0.25%). Therefore the current study results fall under the range of research reports.



The effect of processing on treatable acidity is highly variable. While boiling and soaking might lead to a decrease due to leaching, roasting could potentially increase it due to concentration. To know the specific impact on *Mucuna pruriens* seeds, direct experimental analysis of treatable acids in raw versus processed samples would be necessary. The pH data provided in the abstract (relatively stable) doesn't directly indicate changes in the total acidic content measured by titration.

The allelophatic effect: The allelophatic effect **of** *mucuna prurience* on haricot bean and coffee was evaluated by observing the growth performance of both crops. The result showed that, the growth performance of intercropped coffee and haricot bean is very nice and impressive. From observation, the morphology and anatomy of both crops showed that there is positive effect of *mucuna prurience* on those crops by nitrogen fixation b /c it is legumnes crop.

5. CONCLUSION AND RECOMMENDATION

6. Conclusion

The qualitative phytochemical analysis of the plant extract revealed the presence of several bioactive compounds, including alkaloids, flavonoids, phenolic compounds, and saponins. These findings suggest that the plant extract possesses a diverse range of potential medicinal properties. Further studies are needed to isolate and identify the specific compounds responsible for the observed activities and to evaluate their potential therapeutic applications. This study also provides a detailed analysis of the impact of boiling, roasting, and soaking on the nutritional and physicochemical properties of the food matrix, highlighting the range of changes observed within each treatment and comparing these findings with existing literature. Roasting effectively reduced moisture and concentrated fat, while boiling and soaking led to an increase in carbohydrate proportion and a reduction in protein and mineral content, consistent with findings in other food systems. The relatively stable pH across treatments suggests that these methods, under the current conditions, do not drastically alter the acidity. These results emphasize the importance of considering the specific processing method and its potential effects on the nutritional profile of food. Future research should focus on quantifying nutrient loss and retention rates during these processes, examining the bioavailability of the altered nutrients, and optimizing processing conditions to enhance nutritional quality.

7. Recommendation

Based on the research results provided, a recommendation can be generated to further explore the potential health benefits and applications of Mucuna pruriens seeds. The phytochemical screening of Mucuna pruriens seeds revealed the presence of various bioactive compounds such as alkaloids, carbohydrates, reducing sugars, glycosides, proteins, amino acids, flavonoids, phenolic compounds, tannins, saponins, terpenoids, and resins. These compounds are associated with diverse biological activities including antioxidant, anti-inflammatory, and antimicrobial properties.

Given the rich phytochemical composition of Mucuna pruriens seeds, it is recommended to conduct further studies to investigate the specific health-promoting properties of these bioactive compounds. Research focusing on the isolation and characterization of individual compounds could provide insights into their potential medicinal applications. Additionally, exploring the synergistic effects of these compounds in Mucuna pruriens seeds could lead to the development of novel therapeutic agents or functional food products.

Furthermore, comparative studies analyzing the phytochemical profiles of Mucuna pruriens seeds from different geographical regions could help identify variations in bioactive compound content. Understanding these variations could contribute to optimizing cultivation practices and enhancing the nutritional and medicinal value of Mucuna pruriens seeds.

In conclusion, the research findings highlight the importance of Mucuna pruriens seeds as a valuable source of bioactive compounds with potential health benefits. Further research in this area could pave the way for the development of new pharmaceuticals, nutraceuticals, or dietary supplements harnessing the bioactive potential of Mucuna pruriens seeds.

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