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Development And Evaluation Of Sprouted Millet Malt Incorporated With Drumstick Seeds Powder To Control Hypertension

[1]S. Divyadharshini [2] Dr. R. Balasasirekha [3]Dr. K. Karthikeyan

1. Research Student, Department of Food Science and Nutrition, Periyar University, Salem -11, Tamil Nadu, India
2. Assistant Professor (senior scale) Department of Food Science and Nutrition, Avinashi lingam Institute for Home Science, Coimbatore-641 043, Tamilnadu.
3. Assistant Professor (senior scale) Department of Food Science and Nutrition, Periyar University, Salem -11, Tamil Nadu, India

Corresponding Author,
S. Divyadharshini,
Research Student, Department of Food Science and Nutrition, Periyar University, Salem -11, Tamil Nadu, India.

Abstract

Hypertension, a major health issue in developing countries, contributes significantly to cardiovascular disease. This study explores the potential of drumstick seed powder incorporated sprouted millet malt in managing high blood pressure. Drumstick seed powder showed moderate toxicity in the brine shrimp assay, while heavy metal levels showed within safe limits and did not contribute toxicity. Quantitative analysis using UV-spectrophotometry. Nutrient analysis showed that drumstick seed powder contained high levels of calcium, magnesium and potassium, along with vitamin C, all of these nutrients help for antihypertensive properties.

The Sorghum Millet was germinated and malted and also drumstick seed powder was incorporated in 5g, 10g and 15g variations. Sensory evaluation using a 9-point hedonic scale, indicated that 10 g of drumstick seed incorporation variation in sprouted sorghum millet malt had the highest acceptability. Phytochemical analysis of SM2 showed the presence of coumarins, flavanoids, phenols and polyphenols. In Quantitative analysis using UV- Spectrophotometery results showed that coumarins were present at 0.12 mg/g, flavonoids at 37.8 mg/g, phenols at 44.3 mg/g, and polyphenols at 25 mg/g. The moisture content, pH, bulk density, tapped density, color and water absorption capacity are confirmed in physicochemical properties.

Keywords: Hypertension, drumstick seed powder, sprouted sorghum millet, phytochemical analysis, nutrient analysis, physicochemical properties and shelf life.

Introduction

The *Moringa oleifera* tree belongs to the *Moringaceae* family and has gained global recognition for its nutritional value and numerous health benefits and is mainly grown in Asia and Africa. These seeds are small, round, and have a unique wing-like structure. It is particularly valued for its high content of vitamins, minerals, and antioxidants, including vitamin C and B vitamins. Additionally, they are rich in essential minerals such as potassium, phosphorus, magnesium, calcium, and iron. The seeds contain significant amounts of protein, including essential amino acids like arginine and serine, as well as healthy fats, particularly monounsaturated fatty acids like oleic acid (Bhuvaneswari G, *et al.*, 2016).

In India, millet cultivation dates back to around 2000 BCE during the Indus Valley Civilization, where pearl millet was a staple crop. Due to their hardy and drought-resistant nature, millets adapted well to India's diverse climatic conditions. Millets comprise a group of small-seeded grasses, including pearl millet, finger millet, sorghum, foxtail millet, proso millet, little millet, and kodo millet (Zeenat A, *et al.*, 2021). Sprouting millet grains leads to significant physical, biochemical, and nutritional changes that enhance their overall quality. During sprouting, the grains absorb water, swell, and soften, resulting in increased moisture content and weight. This process enhances nutrient digestibility, making it essential in human nutrition. Additionally, some sprouted millet flours exhibited potential for use in the food industry due to their enhanced functional properties (Panda D, *et al.*, 2019).

Sorghum bicolor (L.) Moench, belongs to the botanical family *Poaceae* was first described by Pliny (60-70 A.D.), initially classified under the genus Milium, followed by multiple reclassifications before being assigned to the genus Sorghum in the early 20th century (David O G, *et al.*, 2022). Germination increases essential amino acids crucial for protein synthesis in the human body. During sprouting, enzymatic activity breaks down complex macronutrients, reducing antinutritional factors like tannins and phytic acid while increasing mineral content (calcium, iron, magnesium, and potassium) and vitamins, particularly B- complex and vitamin C. Sprouting also enhances flavor and taste while promoting gut health through increased dietary fiber (Saithalavi K. M., *et al.*, 2021).

Sorghum is also a rich source of phytochemicals, including anthocyanins, phenolic acids, and sterols. Red sorghum contains higher anthocyanin levels, while white sorghum has lower anthocyanins but still provides significant antioxidant, anticancer, antimicrobial, and immune-boosting properties. Germination improves the nutritional profile by breaking down complex compounds, reducing antinutritional factors, and increasing total free amino acids, polyphenols, and antioxidant activity (Hassan S., et al., 2020).

Hypertension is a growing public health concern worldwide, often linked to poor dietary habits and lifestyle modifications. Functional foods that support cardiovascular health, hypertension management, and diabetes are in increasing demand. This study explores the development of a millet malt incorporated with *Moringa* seed powder, aiming to assess its nutritional, phytochemical, and antihypertensive potential (Katherine T M, *et al.*, 2020).

Objective

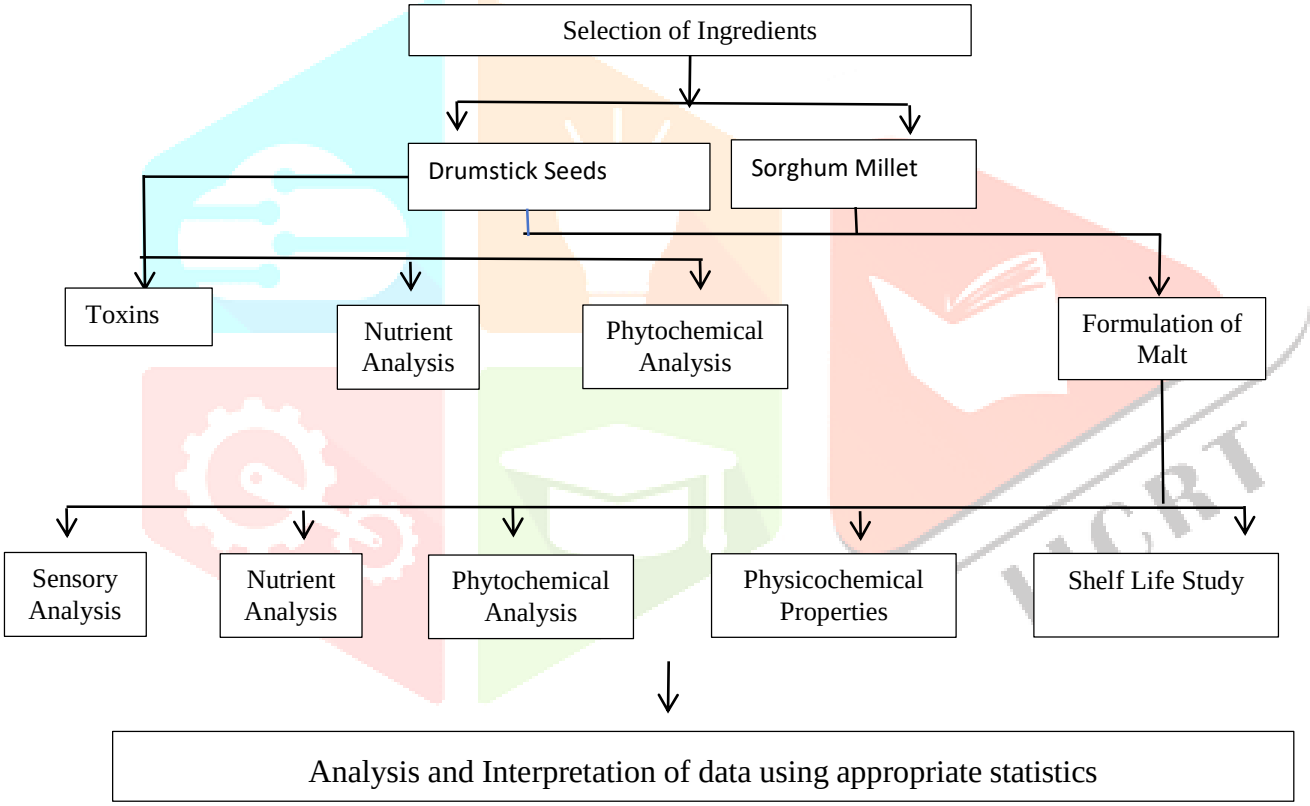
The **Primary Objective** of the study is

- Identifying the potentials of drumstick seed powder incorporated sprouted sorghum millet malt in managing hypertension.

The **Secondary Objective** of the study is,

- Identify the toxins, nutrients and phytochemicals present in drumstick seeds.
- Development and standardisation of sprouted sorghum millet malt using drumstick seeds.
- Conduct sensory analysis to identify the best variation.
- Evaluate the nutrients and phytochemicals present in the best accepted variation.
- Analyse the physicochemical properties of the millet malt.
- Conduct the stability and shelf-life studies of the millet malt.

Methodology



Materials and Methods

Procurement of Raw Ingredients

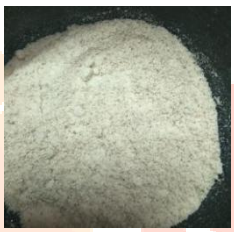
Ingredients required for the standardization of recipes was procured from Super Market, Saibaba Colony in Coimbatore. 1 kg each of sorghum is the essential ingredients used in product development. Additionally, 100g each of other ingredients namely cashew nuts, almonds, makhana and pumpkin seeds were procured. Flavouring ingredients that is used to enhance the flavour of the product namely 50g of rosemary and 50g of cardamom were procured.

Preparation of Drumstick Seed Powder

Drumstick seeds were taken from the kitchen garden of the households in Chitoor of Salem District. The seeds from the drumstick was separated carefully from the mature pods which turn brown and start to split open, revealing the seeds inside. Once the pods are dried and split, carefully removed the seeds from the pods. A knife was used to cut the pods open and removed the seeds. The weight of the drumstick seeds taken from the pod was 250 grams. The drumstick seeds are rinsed thoroughly under running water to remove any dirt or debris. Drained the water and spread the cleaned seeds on a clean cotton towel to remove excess moisture. Allowed them to air dry completely in a well-ventilated area. This may take two days to remove moisture depending on humidity levels and allowed to completely dry and until crisp. It took 5 days for the drumstick seeds to shade dry. Transferred the dried seeds into a high-powered blender. Ground the seeds until they form a fine powder. The weight of the ground powder was 232 grams. Sieved and allowed the ground drumstick seeds powder to cool completely before storing. Stored the powder in an airtight container in a cool and dry place.

Drumstick Seeds

Drumstick Seed Powder



Conduct of Toxicity Studies in Drumstick Seeds

Toxicity studies are used to investigate the safety compound. It is to check whether the substance is safe for humans, animals and environment.

Brine Shrimp Assay - Brine Shrimp Assay toxicity study was conducted to find toxins present in drumstick seeds. The extract of drumstick seeds was prepared by aqueous solutions.This test helps to assess the cytotoxicity, which can be indicated by potential bioactivity. Hatch the Artemia salina eggs in artificial seawater 28 - 30°C. After hatching collected the active nauplii (larvae) used for the experiment. Dissolve the *Moringa Oleifera* seed extract in appropriate solvent. Concentrations used for the extract are 10, 50, 100, 500 and 1000 µg/ml. Dispence 20 - 30 nauplii in each test tube at different concentations. Incubate 24 hours at room temperature.Count the number of dead and surviving nauplii under the microscope. Calculate the percentage mortality concentration (Rowland O U, et al., 2015).

Heavy Metal Analysis for Drumstick Seeds - Heavy metals are considered harmful to living organisms due to their ability to accumulate in specific body tissues. Their buildup is linked to a range of health issues, including neurological, kidney-related, cancer and immune system disorders. The heavy metals namely mercury, lead, cadmium and arsenic was analysed in drumstick seeds using standard procedures as given by AOAC, 2005.

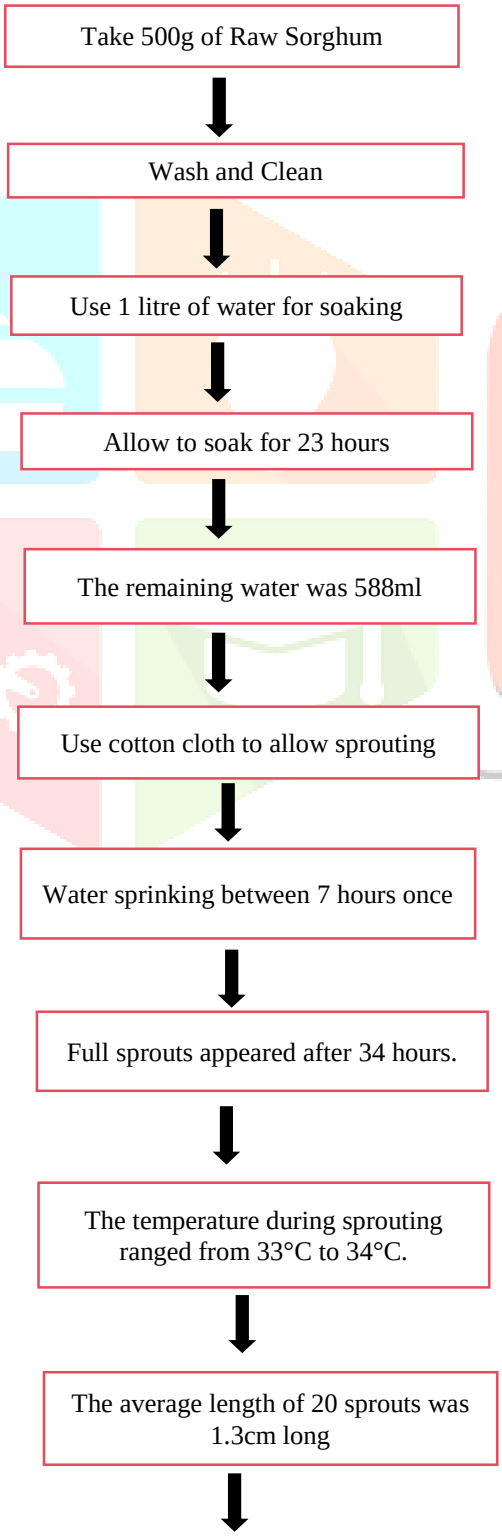
Standardization of Millet Sprouts

The millets selected and included in the current study are sorghum, finger millet, and pearl millet.

TABLE - I

Millets	Initial Weight (g)	Water used for Soaking (ml)	Hours of Soaking (hrs)	Remaining Water (ml)	Hours of Sprouting (hrs)	Final Weight of Sprouted Millet (g)	Final Weight of Millet Powder (g)
Sorghum	500	1000	23	588	34	513.9	502.1

The flow chart presents the Processing and preparation of Sprouted Sorghum



Allowed to shade dry for 15 hours

↓

The weight of the dried sorghum was 513.9g.

↓

Final weight of sprouted sorghum millet flour was 502.1g.

Sorghum Grain

Soaking in water

Sprouted Sorghum

Measuring Sprouts Length

Drying sprouted Sorghum

Sprouted Sorghum Powder

Formulation and variation of Drumstick Seed Powder Incorporated with Sprouted Sorghum Millet Malt

The nutrient-rich millet malt was formulated and standardised using ingredients in various proportions, and the best variation was selected. The selected ingredients was measured according to the specified variations.

TABLE - II
DEVELOPMENT OF SPROUTED SORGHUM MALT INCORPORATED WITH DRUMSTICK SEED POWDER (in g)

INGREDIENTS	VARIATION 1 SM1 (g)	VARIATION 2 SM2 (g)	VARIATION 3 SM3 (g)
Sprouted Sorghum flour	40	40	40
Drumstick Seeds powder	5	10	15
Cashew Nut flour	15	15	15
Almond Flour	10	15	10
Pumpkin Seeds flour	15	15	15
Makhana Flour	15	15	10

Sensory Evaluation of Millet Malt

Sensory evaluation is the scientific discipline used to evoke, measure, analyse and interpret human reactions to those characteristics of food and beverages as they are perceived by the senses of sight, smell, taste and touch. The nine-point hedonic scale used in the consumer's overall liking assessment includes nine categories: 1 (dislike extremely), 2 (dislike very much), 3 (dislike moderately), 4 (dislike

slightly), 5 (neither like nor dislike), 6 (like slightly), 7 (like moderately), 8 (like very much), and 9 (like extremely) (Catherine, C., *et al.*, 2024).

Phytochemical Analysis of Drumstick Seeds and the Best Accepted Sprouted Sorghum Millet Malt Variation

Weighed 10g of drumstick seeds powder and extracted using aqueous extraction, which is the most commonly used method for drumstick seeds. The best-accepted millet malt was extracted using aqueous solution. Ten grams each of the drumstick seed powder and the best-accepted sprouted sorghum millet malt variation was weighed separately and allowed to soak in distilled water for 24 hours. The extract is then filtered and concentrated for further analysis. The obtained aqueous extract is subjected to phytochemical screening to identify bioactive compounds such as alkaloids, flavonoids, tannins, saponins, steroids, terpenoids, and glycosides.

Nutrient Analysis for Drumstick Seeds and the best accepted Sprouted Sorghum Millet Malt Variation

Nutrient analysis is the process of determining the nutritional composition of food or plant samples, including key nutrients such as proteins, carbohydrates, fats, vitamins, minerals, and fiber (Dimitrova, M., 2019). Nutrient analysis of drumstick seeds includes protein, vitamin C, calcium, phosphorus, potassium, and magnesium. The nutrient analysis of the best accepted sprouted sorghum millet malt includes carbohydrates, protein, fat, vitamin C, calcium, phosphorus, potassium, and magnesium. The procedure used for the analysis followed by AOAC, 2002 guidelines.

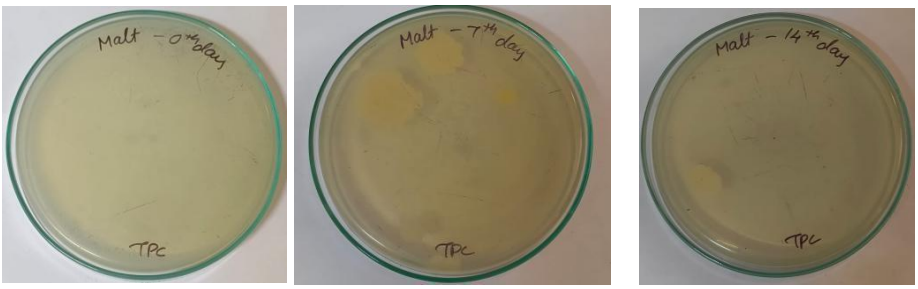
Physicochemical Analysis of the Sprouted Sorghum Millet Malt

Physicochemical analysis of malt involves examining its physical and chemical properties, such as moisture content, pH, tapped density, bulk density, color and water absorption capacity using standard procedures. These properties are essential for determining the malt's quality, consistency, and suitability in brewing and food production. .

Conduct of Shelf Life studies of the Sprouted Sorghum Millet Malt

The shelf life of malt refers to the period during which the product retains its quality, nutritional value, and safety for consumption under proper storage conditions. The total plate count was analysed on 1st , 7th and 14th day to identify the growth of micro organisms using standard procedures.

0^{th} Day 7^{th} Day 14^{th} Day



Consolidation, Statistical analysis and Interpretation of data

The data was compiled and analyzed using statistical methods. Using suitable statistical method, mean acceptability score was computed. The collected data was systematically analyzed to draw conclusions.

Results and Discussion

Toxic Substances in Drumstick seeds

The toxicity analysis was carried out to assess the safety and potential adverse effects of the test sample. Through this evaluation, it was determined whether any harmful properties that could impact health were present in the sample.

Brine Shrimp Assay

Table III presents the toxicity assessment of the Drumstick Seed Powder sample conducted using Brine Shrimp Assay.

TABLE III
BRINE SHRIMP ASSAY OF DRUMSTICK SEED POWDER

Sample Code	Concentration (µg/ml)	Mortality of Brine Shrimp (no. of shrimps death)					
		1	2	4	6	24	% Mortality (24 h)
Drumstick Seed Powder (DS1)	100	0	2	2	4	7	23
	250	1	3	4	4	7	23
	500	1	1	2	5	9	30
	1000	0	0	1	4	9	30
	1500	1	2	2	4	9	30
Control K2Cr2O7	1(mg/ml)	30	-	-	-	-	100
Blank	Saline Water	0	0	0	0	0	0

The Brine Shrimp Assay was performed at concentrations ranging from 100 to 1500 µg/ml, with mortality observed at various time intervals. After 24 hours, 23% mortality was recorded at 100 µg/ml, and full mortality (30 shrimp) occurred at 500 µg/ml and above. The positive control (potassium dichromate) caused 100% mortality at lower concentrations, indicating high toxicity. No mortality was observed in the blank. These results indicate that drumstick seed powder shows moderate toxicity, while potassium dichromate is highly toxic even at minimal doses. (Dhigna, D L, *et al.*,) showed the moderate toxicity in drumstick seed powder in 500-1500 concentrations.

Heavy Metals in Drumstick Seed Powder

The Brine Shrimp Assay indicated moderate toxicity at both low and high concentrations. Therefore, a heavy metal analysis was conducted for this study (Table IV).

TABLE IV
HEAVY METALS TEST IN DRUMSTICK SEED POWDER

Heavy Metals	Result Values(mg/kg)	Test Method
Lead	0.08	BIS/AOAC, 2005
Cadmium	<0.01	
Arsenic	<0.01	
Mercury	<0.01	

The metals in drumstick seed powder analyzed are lead, cadmium, arsenic, and mercury. Lead was detected at a concentration of **0.08** mg/kg, while cadmium, arsenic, and mercury were each found to be

below the detection limit of **0.01** mg/kg. These findings suggest that the levels of heavy metals present in the drumstick seed powder are within permissible limits and do not contribute to toxicity. Hence can be incorporated into the food safety for consumption.

Sensory Scores and Acceptability of the developed Millet Malt

a) Sprouted Sorghum Malt

The sensory attributes of sprouted sorghum millet malt was evaluated by semi-trained panel members and is presented in Table V.

TABLE V
SENSORY SCORES OF SPROUTED SORGHUM MILLET MALT

n=30

Criteria	Standard SMS	Variation 1 SM1	Variation 2 SM2	Variation 3 SM3
Appearance	8.5±1.0	8.4±0.8	8.9±0.2	8.3±0.7
Color	8.1±1.1	8.1±0.5	8.9±0.2	7.5±1.07
Texture	8.1±0.9	7.9±0.5	8.8±0.4	6.6±0.9
Flavour	7.7±1.4	7.7±1.1	8.5±1.1	7.03±0.6
Taste	8.1±0.9	7.5±0.8	8.8±0.5	6.4±1.1
Overall Acceptability	8.1±1.06	7.92±0.74	8.78±0.48	7.1±0.8
F Value - Standard Vs Variations		4.41*	13.02*	5.67*

***Significant at 5% level**

Based on the sensory evaluation results, SM2 was the most acceptable variation among the three samples, with an F-test value of 13.02.

Phytochemicals in Drumstick Seeds and best accepted variation of Sprouted Sorghum Millet Malt

Phytochemicals are bioactive compounds primarily produced by plants, serving as key sources of active ingredients in the pharmaceutical industry (Mendoza, N, *et al.*, 2018).

TABLE VI
PHYTOCHEMICALS IN DRUMSTICK SEEDS AND BEST ACCEPTED VARIATION OF MILLET MALT

Metabolite	Test Performed	Drumstick Seeds	Sprouted Sorghum Millet Malt (SM2)
Alkaloids	Mayer's Test	+	-
Coumarins	NaOH	-	+
Flavanoids	HCl	+	+
	H2SO4	-	+
	Alkaline Test	+	-
Phenols	Lead Acetate Test	-	+
Polyphenols	Folin-Ciocalteu method	-	+
	Ferric Cyanide	+	+
Saponins	Foam Test	-	-
Tannins	Ferric Chloride test	+	-

+ Present; - Absent

Aqueous extracts of drumstick seeds and best-accepted sprouted sorghum millet malt variation SM2 was tested for phytochemicals. Drumstick seeds showed the presence of alkaloids, flavonoids, polyphenols, and tannins, with absence of phenols and saponins. SM2 contained coumarins, flavonoids, phenols, and polyphenols.

a) Quantitative Analysis of Drumstick Seeds and best accepted Millet Malt

To evaluate the presence of phytochemicals in the selected samples, a quantitative analysis was conducted using the UV-VIS spectrophotometric method. Tables VII present the results of the quantitative analysis conducted on drumstick seed powder and the best-accepted millet malt formulation.

TABLE VII

PHYTOCHEMICAL (QUANTITATIVE) IN DRUMSTICK SEED POWDER AND BEST ACCEPTED SPROUTED SORGHUM MILLET MALT

Phytochemicals	Drumstick Seed Powder (DS) (mg/100g)	Sprouted sorghum millet malt (SM2) (mg/100g)
Alkaloids	36	-
Coumarins	-	0.12
Flavanoids	24	37.8
Phenols	-	44.3
Polyphenols	-	25
Tannins	47	-

The table compares phytochemical content in drumstick seed powder and best accepted sprouted sorghum millet malt. The aqueous extract of drumstick seeds was used to identify various phytochemicals, including alkaloids, flavonoids, phenols, polyphenols, saponins, and tannins. The best-accepted sprouted sorghum millet malt, i.e., variation 2 (SM2), was analyzed for the presence of phytochemicals using an aqueous extract, as these compounds are easily soluble in water. Phytochemical analysis of SM2 showed the presence of coumarins, flavanoids, phenols and polyphenols, while alkaloids, saponins and tannins were absent as determined by using standard procedures.

Nutrients in Drumstick Seeds and best accepted Sprouted Sorghum Millet Malt

Among the variations developed, the second variation incorporated at 10% of each malt was found to be the most acceptable based on sensory evaluation

TABLE VIII
NUTRIENT ANALYSIS FOR BEST ACCEPTED MILLET MALT

Nutrients	Drumstick Seeds g/100g	Sprouted Sorghum Malt (SM2) g/100g
Carbohydrates (kcal)	-	73.2
Protein (g)	3.5	5.8
Fat (g)	-	3
Vitamin-C (mg)	15.6	67.58
Calcium (mg)	196	487.6
Magnesium (mg)	266	131.78
Potassium (mg)	361	349
Phosphorous (mg)	160	234.87

Carbohydrate - The carbohydrate content of different sprouted millet malts was analyzed to assess their nutritional value. The results showed that sprouted sorghum millet malt contained 73.2 g per 100 g. According to Ikram, et al., (2021), sprouted sorghum millet contains 132 mg of carbohydrates per 100 g, which differs significantly from findings.

Protein - Drumstick seeds powder was found to contain 3.5 g of protein per 100 g. Among the millet malts, sprouted sorghum malt contained 5.8 g of protein per 100 g. In contrast, Ikram, *et al.,* (2021) reported a significantly lower carbohydrate content of 9.9 g per 100 g for sprouted sorghum millet.

Fat - The results showed that sprouted sorghum malt contained 3 g of fat per 100 g, sprouted finger millet malt contained 2.1 g per 100 g. According to published literature, the fat content of sprouted sorghum millet was reported as 3 g/100 g by Ikram *et al.,* (2021),

Vitamin C - It was found that drumstick seeds contained 15.6 mg per 100 g of vitamin C. A slightly lower value of 14.4 mg per 100 g was reported for drumstick seed powder by Rani E. A. et al. (2017). In the present study, the highest vitamin C content was observed in sprouted sorghum millet malt 67.58 mg per 100 g.

Calcium - The results revealed that drumstick seeds contained 196 mg per 100 g of calcium. Among the millet malts, sprouted sorghum millet malt calcium content was 487 mg per 100 g.

Magnesium - The results showed that drumstick seeds contained 266 mg per 100 g, making them the richest source of magnesium among all samples tested. The sprouted sorghum millet malt was 131.78 mg per 100 g.

Phosphorous - The results showed that drumstick seeds contained 361 mg of phosphorus per 100 g. Among the millet malt, sprouted sorghum millet malt contained 349 mg per 100 g.

Potassium - The analysis revealed that drumstick seeds contained 160 mg per 100 g. Sprouted sorghum millet malt had 234.87 mg per 100 g.

Physicochemical properties of best accepted Millet Malt

The physicochemical properties were analyzed for the best accepted sprouted sorghum millet malt.

TABLE IX
PHYSICOCHEMICAL PROPERTIES FOR BEST ACCEPTED SPROUTED SORGHUM MILLET MALT

Physicochemical Properties	Sprouted Sorghum Malt SM2
Moisture Content	5g
pH	6.08
Bulk Density	5.6g/100ml
Tapped Density	11.6g/100ml
Water absorption Capacity	120ml/100g

The physicochemical analysis of SM2 shows a low moisture level (5 g), which helps in extending shelf life and minimizing microbial spoilage. Its pH (6.08) is close to neutral, making it suitable for both consumption and processing. The bulk and tapped density values indicate good handling and packing properties, while the high water absorption capacity (120 ml/100 g) reflects better reconstitution and ease of digestion.

Color

TABLE X
COLOR FOR BEST ACCEPTED MILLET MALT

Sample	L*	a*	b*
Sprouted Sorghum Malt (SM2)	22.6	3.38	19.11

L*-Lightness/Darkness; a*-Red/Green; b*-Yellow/Blue

The color analysis of the best-accepted sprouted sorghum millet malts showed that highest L* (22.6), a* (3.38), and b* (19.11) values, indicating a lighter and more vibrant color.

Shelf life of the Millet Malt

Refrigeration Storage - The best accepted sprouted millet malts, coded as SM2 (sprouted sorghum malt) was stored under refrigeration conditions at 10°C for a period of 15 days. No microbial growth was observed, and sensory qualities like taste, texture, flavor, and appearance remained stable. Refrigeration effectively preserved quality, confirming malt are suitable for extended shelf life.

a) Room Temperature - The best accepted variations of sprouted sorghum millet malt (SM2) was selected for storage studies. These samples was stored at room temperature, maintained between 30–35°C, reflecting the average climatic conditions in Coimbatore during the 15 days study period. SM2 showed slight texture and flavor changes. Proper packaging is essential for shelf life. Generally, millet malt can last up to 18 months when stored at 25°C and 30% humidity.

Total Plate Count - A shelf life study was conducted for the best accepted sprouted sorghum millet malt, and microbial testing was performed on the samples at 14-day intervals.

TABLE XI
TOTAL PLATE COUNT FOR BEST ACCEPTED MILLET MALT

Sample	RESULTS (CFU/g)		
	0th Day	7th Day	14th Day
Sprouted Sorghum Millet Malt (SM2)	3x101	7x101	11x101

The microbial analysis of sprouted sorghum millet malt over 14 days showed a steady increase in microbial load in sample. Sorghum Millet showed better microbial stability with the lowest final count (11×10^1 CFU/g).

Conclusion

The study developed millet malt formulations by incorporating drumstick seed powder to manage hypertension. Drumstick seed powder was found to be nutritionally rich, moderately toxic at high doses, and safe based on heavy metal analysis. GC-MS confirmed bioactive compounds like unsaturated fatty acids, plant sterols, and phenolics. The sprouted sorghum millet malt have 10g drumstick powder variation was most acceptable based on sensory evaluation. Nutritional and phytochemical analyses showed that SM2 had high calcium and vitamin C. Physicochemical analysis indicated SM2 had the best bulk density and color. Microbial studies revealed that SM2 had the best shelf stability. Overall, SM2 (sprouted sorghum malt with 10g drumstick seed powder) was identified as the most suitable formulation for managing hypertension due to its rich nutrient profile, bioactive content, sensory acceptability, and shelf life.

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