

# Evaluation of Nutrient, Mineral analysis and quality characterization of *Gymnema sylvestre* multi grain cookies for diabetes

## Abstract

Diabetes is a group of metabolic disorders with various etiologies that are defined by persistent hyperglycemia and worsening carbohydrate, lipid, and protein digestion due to flaws in insulin discharge and insulin activity. Indians account for one out of every five diabetics of the world. Around 35 million Indians suffer from diabetes, which affects around 150 million individuals worldwide. Because health is a major concern in people's daily lives, foods that are healthy and beneficial to health are becoming increasingly popular. Natural food variations are widely used for the prevention and treatment of a variety of healthy issues. Supplements such as protein, iron, and calcium can be found in plenty of treats. The goal of this study was to develop solid treats using *Gymnema sylvestre* leaf powder, an Ayurvedic component with increased nutraceutical value. Multigrain powder, heating powder, palm sugar, cardamom powder, Margarine, salt, bubbling blend, and *G.sylvestre* leaf powder were among the ingredients used to make the solid snacks. The powder was created by drying the leaves in a plate dryer at 50°C for 2 hours before crushing in a home processor. *G.sylvestre* leaf powder was fused at different percentages: 0.25 percent, 0.50 percent, 0.75 percent, and 1.00 percent. Supplement testing, mineral analysis, and tactile testing are not set in stone. Atomic Absorption Spectroscopy was used to determine mineral composition. Over a variety of examples, test 0.50 percent sound snacks were found to be organoleptically superior.

**Keywords:** Cookies, Nutritional analysis, Mineral analysis, Microbial analysis, *G.sylvestre*.

## Introduction

Diabetes mellitus is a metabolic disorder characterised by a lack of the chemical insulin in the blood, resulting in abnormalities in the body's absorption of carbs. In view of the fact that the age of onset isn't the underlying predictor of the type of diabetes, the phrases 'adolescent onset' and 'developmental onset diabetes have been replaced with 'Type I and Type II' [1]. Heredity, incorrect dietary proclivities, lack of movement, the effects of pharmaceuticals and poisons, the impact of chemicals, mental factors, and diabetes as a complication of other illnesses such as pancreatitis and cardiovascular failures are the most generally acknowledged causes of diabetes [2]. Polydipsia, polyuria, and polyphagia are the

34 most common symptoms of diabetes mellitus. Vision blurring, skin irritation, general  
35 weakness, and a lack of solidarity are some of the negative effects. Finally, it causes water  
36 and electrolyte imbalances, ketoacidosis, and a trance-like state [3]. Retinopathy,  
37 nephropathy, neuropathy, and repeating myocardial dead tissue are among the persistent  
38 problems, which are associated with an increase in the frequency of congestive  
39 cardiovascular breakdown, ulceration, foot infection, and even gangrene [4]. For diabetics, a  
40 high-complex-sugar, low-fat diet that includes a variety of soil-based products would be ideal  
41 [5].

42 One of the nibbling things, a treat, contains refined flour, sugar, and spread at first.  
43 Because health is such a major concern in our daily lives, nutritious and beneficial things are  
44 in high demand. Treats are the most well-known bread shop items due to their excellent  
45 nutritional value, ready-to-eat nature, and easy availability in a variety of shapes and sizes at  
46 a fair price. Treats are popular because they are high in carbohydrates, lipids, and calories.  
47 Currently, the fusion of a few elements in treats has evolved to improve its nutritive and  
48 beneficial qualities. Among other things, treats are known to be a good source of energy,  
49 protein, iron, calcium, and calories, among other things. Treats are not considered to be staple  
50 foods like bread, but they can be considered fibre transporters because of their longer shelf  
51 life, allowing for a wider range of manufacturing and distribution [6]. Margarine is made  
52 from vegetable oils, therefore it contains polyunsaturated and monounsaturated fats, as well  
53 as unsaturated "excellent" fats. When filling in for submerged fat, these fats aid to lower low-  
54 thickness lipoprotein (LDL), or "bad," cholesterol. Cookies' useful features can be enhanced  
55 by altering and supplementing them with health-promoting ingredients such as "Gulmar"  
56 leaves, mixed sugar (fake + regular) sugars, fat substitutes, and wheat flour [7].

57 *G. sylvestre* is a prominent Indian medicinal plant that is widely used in the treatment  
58 of diabetes mellitus. *G. sylvestre* is a basic therapeutic woody climber that belongs to the  
59 Asclepiadaceae ('Milk Weed Family') family. 'Wonder natural product' is one of the unique  
60 names for this plant species. The name '*Gymnema sylvestre*' comes from a Latin word that  
61 means 'bare' and, which means 'from the woods' [8]. *G.sylvestre* can be found in India,  
62 Malaysia, Sri Lanka, Australia, Indonesia, Japan, Vietnam, tropical Africa, and western  
63 China in general [9]. *G.sylvestre* is thought to have a potential anti-diabetic effect. When you  
64 bite on the *Gymnema* leaves, the lack of capacity tastes good. Because of qualities that lower  
65 and balance glucose levels, concentrates of its leaves and roots are used as a typical treatment  
66 for diabetes in India and parts of Asia. Antimicrobial, antihyphal, antihyper - cholesterolemic,  
67 and hepato-defensive exercises are also performed by the plant. It also acts as a repellent to

68 the caterpillar *prodenia eridania*, prevents dental cavities caused by *Streptococcus mutans*,  
 69 and is used in cosmetics. *Gymnemic* corrosive is found in *G.sylvestre* leaves, and it is known  
 70 to suffocate the glucose transporter from the digestive system to the circulatory system. It  
 71 could potentially restore or regenerate the pancreas' insulin-producing beta cells, as well as  
 72 reduce cholesterol and glucose absorption from the GI tract. Supplementing with *G.sylvestre*  
 73 appears to enhance glucose control in type 2 diabetic patients. Reduced postprandial blood  
 74 glucose resulted in a decrease in HbA1C, lowering the risk of diabetic complications [10].  
 75 This plant's leaves create a zero-calorie, non-nutritive, high-power sugar that can be used as a  
 76 sucrose alternative. Sorbitol, one of the polyols, is a good sugar substitute that has no effect  
 77 on the quality of the treat. For diabetics, sorbitol is preferable to sorbitol as a sugar alternative  
 78 to a variety of foods [7].

79

## 80 **Materials and Methods**

### 81 **Raw material preparation**

82 Multigrain and *G. sylvestre* leaves were purchased from the local market and  
 83 necessary pre -treatments such as washing, drying, grading, sorting, incorporation etc. was  
 84 carried out. Palm sugar, margarine, baking powder and essence were purchased from local  
 85 market. Margarine was stored in the refrigerator at -40°C.

86

### 87 **Cookies Preparation and optimization**

88

**Table 1: Optimization for control and multigrain cookies**

Composition	Control	0.25%	0.50%	0.75%	1%
Multi Grain Powder (Cup)	1	1	1	1	1
Baking Powder (Tablespoon)	1/4	¼	1/4	¼	1/4
Palm Sugar (Cup)	1/4	¼	1/4	¼	1/4
Cardamom Powder (Tablespoon)	1/2	½	1/2	½	1/2
Margarine (Cup)	1/4	¼	1/4	¼	1/4
Salt (Tablespoon)	1/8	1/8	1/8	1/8	1/8

Boiled Mix (Tablespoon)	2	2	2	2	2
<i>G. sylvestre</i> Leaf Powder (%)	-	0.25	0.50	0.75	1

89 Cookies were prepared with some modifications. For the optimization of multigrain  
90 powder, a varying proportion of margarine and palm sugar free sweetener by varying  
91 proportion of two was carried out.

92

### 93 **Proximate composition**

94 Proximate composition (moisture content, carbohydrate, ash, fat, protein, fibre content  
95 and sugar content) was determined by using different methods [11,12,13].

96

### 97 **Mineral determination**

98 The minerals such as sodium and potassium content of sample were determined by  
99 using the Atomic Absorption Spectroscopy as described by [6].

100

### 101 **Determination of metals**

102 All the atomic measurements are carried out with PerkinElmer model 400/HGA  
103 900/AS 800 coupled with Mercury Hydride System-15 (MHS-15) and Flame Photometer.  
104 The Hallow cathode lamps (HCL) for Na and K analyses are used as a light source to provide  
105 specific wavelength of the elements to be determined and high purity (99.999 %) Acetylene  
106 and Nitrous oxide are used to provide constant thermal energy for atomization process and  
107 Argon gas used for carrier gas removal purposes for Graphite furnace.

108

### 109 **Atomic: Absorption Spectrophotometer (AAS)**

110 After calibrating the instrument with prepared working standard, the 10 ml of digested  
111 liquid sample is pipette out to a specific container of Mercury Hydride system analyzer  
112 followed by adding 1.5% Hcl of 10ml as diluents for each flask and blank, 3 % of NaBH<sub>4</sub>  
113 solution in 1% of NaOH is run through the reaction flask to quartz cell without heating  
114 against the calibration curve obtained from concentration vs. absorbance of the prepared  
115 known concentration on the day of the analysis.

116

### 117 **Organoleptic analysis**

118 Sensory evaluation was carried out by a panel of ten semi trained panel members.  
119 Hedonic rating test was employed using a 9-point hedonic scale. Sensory parameters such as

120 colour, taste, texture and overall acceptability were evaluated [13]. The following were the  
121 numerical scores assigned: 9: Like extremely 8: Like very much 7: Like moderately 6: Like  
122 slightly 5: Neither like nor dislike 4: Dislike slightly 3: Dislike moderately 2: Dislike very  
123 much 1: Dislike extremely.

124

### 125 **Isolation of microbes**

126 The sample is mixed with 1L pure water to make solution sample. Then this solution  
127 sample was taken for process. The sterile Petri dish was labeled. Afterwards, 1 ml of the  
128 inoculum was evenly spread over the entire surface of the nutrient agar plates until the  
129 medium no longer appears moist. The flaming and spreading for each of the remaining plates  
130 was repeated. The plates were inverted and incubated at room temperature of at least 48  
131 hours. The plate's colonies were found to be statistically valid [14].

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### 133 **Biochemical Tests**

134 The biochemical tests were carried out for different methods such as Gram staining [14],  
135 Methyl red and Voges- Proskauer test [15], Oxidase test [16], Catalase test [16], Indole test  
136 [16], Urease test [16], Nitrate reduction test [16], Glucose fermentation test [16], Motility test  
137 [15].

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### 139 **Statistical analysis**

140 All experiments were carried out in triplicates. Data obtained were analysed by one-  
141 way analysis of variance (ANOVA) and means were compared by Duncan's New Multiple  
142 Range test (SPSS 21.0 version). Differences were considered significant at  $p < 0.05$ .

143

### 144 **Results and Discussion**

145 In light of the relevant scientific literature, the results collected during this inquiry are  
146 addressed under appropriate areas. Raw materials, control cookies, and healthy cookies were  
147 analysed at varied concentrations (0.25 percent, 0.50 percent, 0.75 percent, and 1.00 percent).  
148 The data collected during the experiments are shown in the tables below, along with a  
149 description of the findings. The amount of *G.sylvestre* leaves per kilogramme was determined  
150 using an electronic digital balance with a sensitivity of 0.01 gm [17].

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### 153 **Nutritional Analysis**

154         The nutritional analysis (ash, moisture, fiber, carbohydrates, protein, fat and  
155 sugar) were analyzed for control and different concentration (0.25%, 0.50%, 0.75%  
156 and 1.00%). The ash value was 1.76 %, 1.95 %, 2.05 %, 1.84 % and 1.74 for control,  
157 0.25%, 0.50%, 0.75%, 1.00% cookies samples, respectively. The residue left after all  
158 the moisture has been removed, as well as the organic material (fat, protein, carbs,  
159 vitamins, organic acid, and so on) has been burnt at a temperature of around 500 °C.  
160 This, is referred as ash in food. The mineral content of the original food is often  
161 measured by ash content [18]. The moisture content was found to be 3.65 %, 3.84 %,  
162 4.23 %, 3.97 % and 3.89 % for control, 0.25%, 0.50%, 0.75%, 1.00% cookies  
163 samples, respectively. Because the fresh plant has a short shelf life and is susceptible  
164 to microbial attack, protracted storage would result in deterioration. This encourages  
165 users to store their items in dry state. In the processing, preservation, and storage of  
166 food, moisture content is one of the most important and often used measurements [18].

167         The amount of non-digestible carbohydrate and lignin in a food or plant is  
168 measured as crude fibre. The crude fibre obtained for cookies was found to be 8.72 %,   
169 10.13 %, 10.46 %, 10.33 % and 10.03 % for control, 0.25%, 0.50%, 0.75%, 1.00%  
170 cookies samples, respectively. This low amount is thought to be optimal since it  
171 improves glucose and fat absorption. Although crude fibre improves digestion, too  
172 much of it can produce intestinal discomfort, reduced digestibility, and reduced  
173 nutritional utilisation [19]. Crude fibre is primarily made up of cellulose, with a little  
174 amount of indigestible lignin [18]. The carbohydrate content of healthy cookies were  
175 found to be 62.14 %, 63.77 %, 63.95%, 63.53 % and 63.66 % for control, 0.25%,  
176 0.50%, 0.75%, 1.00% cookies samples, respectively. When compared to the  
177 Recommended Dietary Allowance (RDA) of 130g, the plant is a moderate source of  
178 carbohydrate [20]. The crude protein of healthy cookies were found to be 8.44%,  
179 9.34%, 10.96%, 10.16% and 9.96 % for control, 0.25%, 0.50%, 0.75%, 1.00% cookies  
180 samples, respectively and represented in Table 2 and Figure 1. The RDA for protein is

181 56 grammes for adults weighing 70 kilogrammes and 46 grammes for adults weight  
 182 50 kilogrammes; children may ingest 2 kilogrammes per day [21].

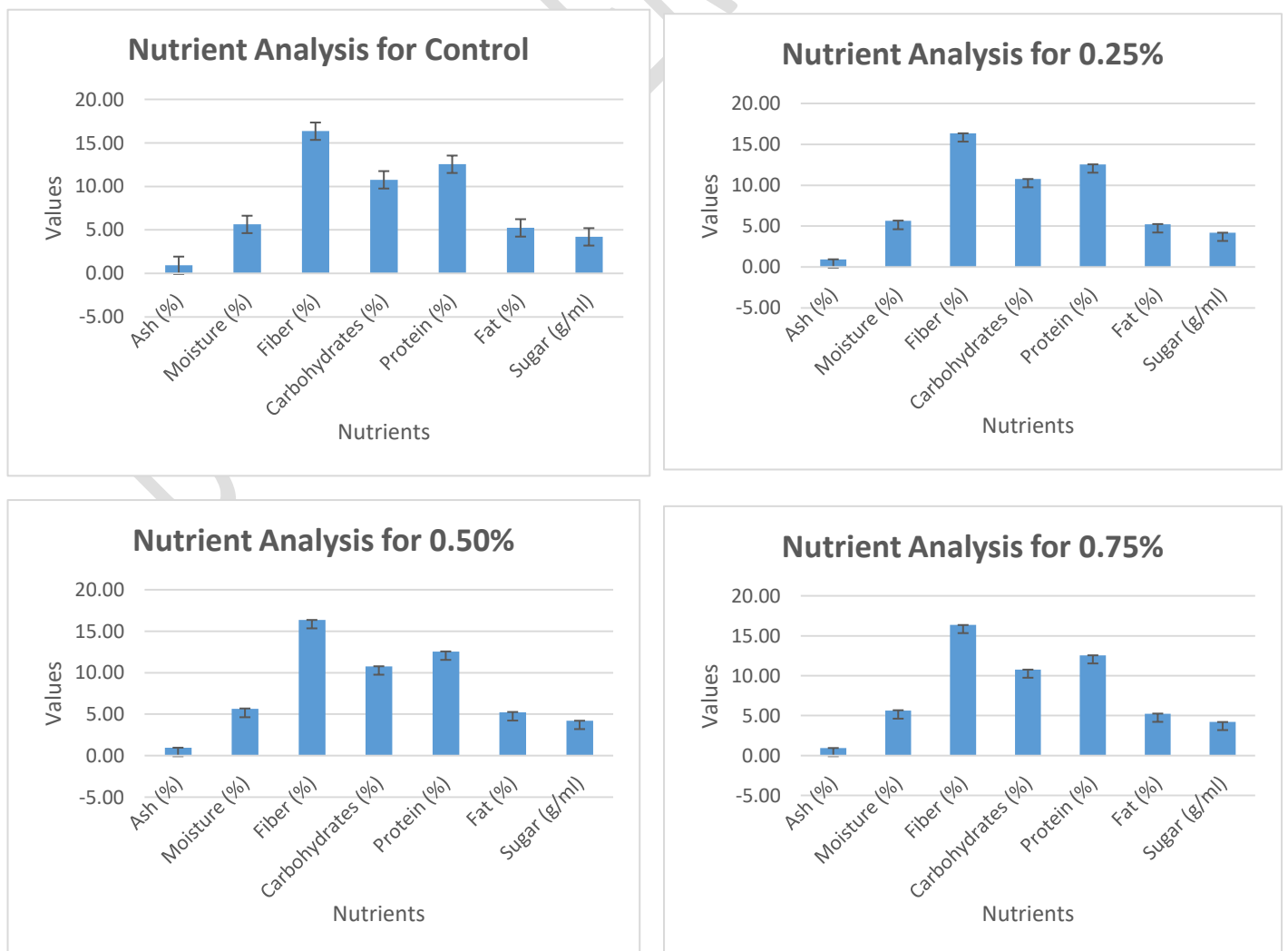
183 **Table 2: Nutrient Analysis for Cookies**

Nutrients Parameters	Control	0.25%	0.50%	0.75%	1.00%
Ash (%)	1.76 ± 0.03	1.95 ± 0.03	2.05 ± 0.02	1.84 ± 0.04	1.74 ± 0.02
Moisture (%)	3.65 ± 0.03	3.84 ± 0.03	4.23 ± 0.02	3.97 ± 0.02	3.89 ± 0.02
Fiber (%)	8.72 ± 0.01	10.13 ± 0.02	10.46 ± 0.02	10.33 ± 0.02	10.03 ± 0.02
Carbohydrates (%)	62.14 ± 0.04	63.77 ± 0.02	63.95 ± 0.04	63.53 ± 0.02	63.66 ± 0.03
Protein (%)	8.44 ± 0.01	9.34 ± 0.03	10.96 ± 0.02	10.16 ± 0.02	9.96 ± 0.03
Fat (%)	21.25 ± 0.03	21.04 ± 0.03	20.97 ± 0.03	19.34 ± 0.03	19.23 ± 0.02
Sugar (g/ml)	18.44 ± 0.03	11.25 ± 0.03	11.36 ± 0.02	11.06 ± 0.03	10.96 ± 0.02

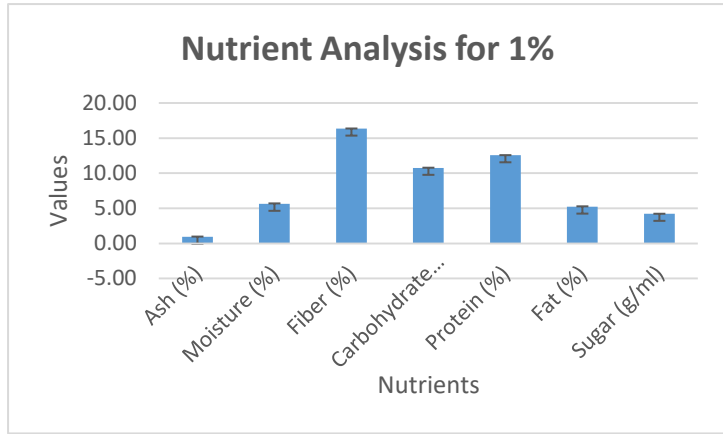
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**Figure 1: Nutrient Analysis for Cookies**



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The plant has a moderate protein content. Plant-based proteins have a lesser nutritional value, according to [20], but when combined with many other protein sources, such as animal protein, they can provide appropriate nutritious value. Protein is required for a variety of bodily processes, including body development, fluid balance, hormone production, enzyme production, and maintaining a robust immune system [22]. The fat content of healthy cookies were found to be 21.25%, 21.04 %, 20.97 %, 19.34 % and 19.23 % for control, 0.25%, 0.50%, 0.75%, 1.00% cookies samples, respectively. The low fat content of 1.83mg/100g is lower than the range (8.3 percent -27.0 percent) observed for various Nigerian green vegetables [23]. Carr's Index and Hausner's ratio for *G. sylvestre* were 20.37 and 1.255, respectively, showing fair compressibility. However, [11,17] obtained similar results for Bulk Density, Tapped Density, Carr's Index, and Hausner's Ratio. The results of proximate composition revealed that multigrain is a good source of carbohydrate, protein, and crude fibre, while *G. sylvestre* powder is a good source of carbohydrate, crude fibre, and crude protein. Moisture content in multigrain and *G. sylvestre* powder was found to be higher while fat content was lower as concentration increased. Multigrain and *G. sylvestre* powder contained higher amount of carbohydrate in 0.50% ( $63.95 \pm 0.04$ ) when compared to other concentrations. Crude fibre, protein and ash content of wheat flour and *G. sylvestre* powder were found to be 10.46, 10.96 and 2.05 per cent respectively.

212 **Mineral analysis**

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Minerals are inorganic elements that serve as structural components and process regulators in the body. The sodium and potassium content of multigrain and *G. sylvestre* powder are shown in Table 3 and Figure 2. Table 3 shows the mineral contents of the healthy

216 cookie samples. The sodium content in healthy cookies were found to be 53.16 mg, 57.51  
 217 mg, 61.25 mg, 59.86 mg and 58.73 mg for control, 0.25%, 0.50%, 0.75%, 1.00% cookies  
 218 samples, respectively. Sodium is a vital mineral that aids in the transmission of nerve  
 219 impulses as well as the preservation of cell osmotic balance. The Recommended Daily  
 220 Allowance for sodium for humans is 1100-3300 mg/100g, according to the National Research  
 221 Council [24]. Dehydration or muscle cramps can result from a sodium deficiency [25]. The  
 222 potassium was the most abundant element in the healthy cookie samples. The highest  
 223 potassium content (257.95 mg) was recorded in 0.50% healthy cookie sample. The potassium  
 224 content in healthy cookies was found to be 173.44 mg, 238.15 mg, 257.95 mg, 241.24 mg  
 225 and 231.65 mg for control, 0.25%, 0.50%, 0.75%, 1.00% cookies samples, respectively.  
 226 According to the National Research Council [24], the Recommended Dietary Allowance for  
 227 potassium for adults is 1875-5625 mg/kg. Potassium is essential for the management of water  
 228 and electrolyte balance and acid-base balance in the body, as well as nerve action and  
 229 muscular function. Potassium deficiency causes muscle paralysis [26]. The sensory analysis  
 230 was found to be good in every of the examined parameters. A panel of ten semi-trained  
 231 judges evaluated the control and healthy cookies for sensory qualities using a 9 point Hedonic  
 232 scale method. Several parameters such physical appearance, texture/mouth feel, taste, colour,  
 233 and overall acceptability were tested. The quality was assessed using the mean values of ten  
 234 semi-trained assessors. Sensory evaluation revealed that the control sample cookies 0.50  
 235 percent scored well on all metrics when compared to the others. As a result, sample 0.50  
 236 percent was chosen for the creation of nutritious cookies.

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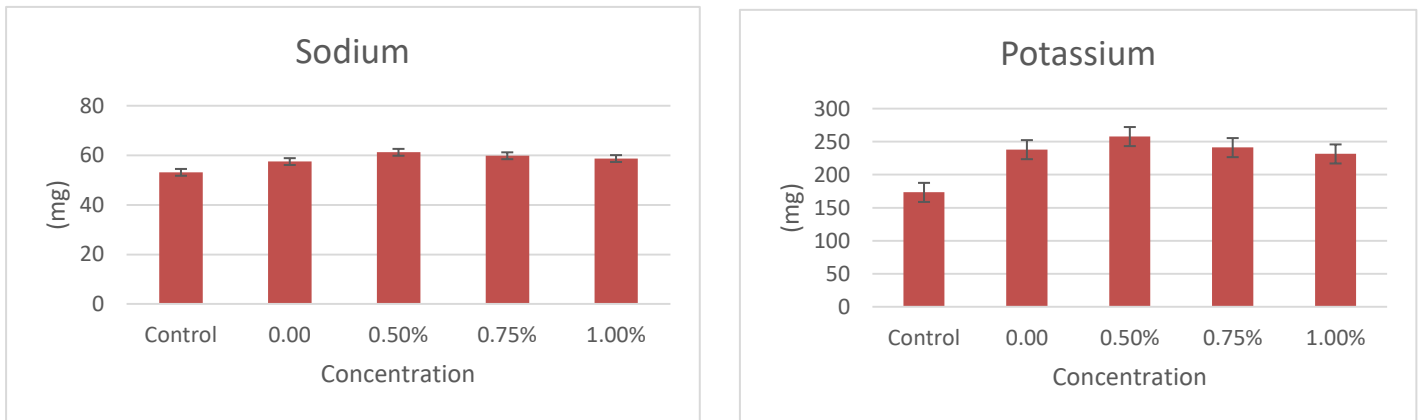
238 **Table 3: Mineral Analysis for Control Cookies**

Minerals	Control	0.25%	0.50%	0.75%	1.00%
Sodium (mg)	53.16 ± 0.02	57.51 ± 0.04	61.25 ± 0.02	59.86 ± 0.02	58.73 ± 0.03
Potassium (mg)	173.44 ± 0.04	238.15 ± 0.02	257.95 ± 0.02	241.24 ± 0.03	231.65 ± 0.02

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240

**Figure 2: Mineral Analysis for Cookies**



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### 242 **Microbial Analysis**

243 The isolation of microbes was carried out in 0.50% cookies. In each and every test  
244 which was performed the results were good in 0.50% cookies. The healthy cookies of 0.50%  
245 concentration were tested in different storages that is AF (Aluminium Foil), LDPE (Low  
246 density polyethylene) and HDPE (High density polyethylene). The CFU count was  $0.12 \times 10^2$   
247  $10^2$ ,  $0.48 \times 10^2$  and  $0.72 \times 10^2$  in day 1, day 15 and day 30 AF samples. However, a higher  
248 CFU count was obtained in HDPE samples ( $0.36 \times 10^2$ ,  $1.98 \times 10^2$  and  $2.52 \times 10^2$  in day 1,  
249 day 15 and day 30 AF samples, respectively) and it is represented in Table 4 and Figure 3, 4  
250 and 5. The dilution was  $10^{-6}$ . The decrease observed in bacterial load after 48 h liquid  
251 fermentation may be as a result of some bioactive substances which might have produced an  
252 inhibitory effect on other organisms involved in the fermentation. This is in line with the  
253 report of Ouoba and Kalui [27,28,29]. Low microbial growth observed after 48 hrs of solid  
254 fermentation may also be as a result of reduction in the moisture content, which is known to  
255 be the most important factor in microbial growth [30,31].

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262 **Table 4: Counts for Colony Forming Unit in 0.50% Cookies**

Samples	Day 1		Day 15		Day 30		Dilution
	No. of Colonies	CFU (No. of Cells/ml)	No. of Colonies	CFU (No. of Cells/ml)	No. of Colonies	CFU (No. of Cells/ml)	
AF	2	$0.12 \times 10^2$	8	$0.48 \times 10^2$	12	$0.72 \times 10^2$	$10^{-6}$
LDPE	7	$0.42 \times 10^2$	36	$2.16 \times 10^2$	46	$2.76 \times 10^2$	$10^{-6}$
HDPE	6	$0.36 \times 10^2$	33	$1.98 \times 10^2$	42	$2.52 \times 10^2$	$10^{-6}$

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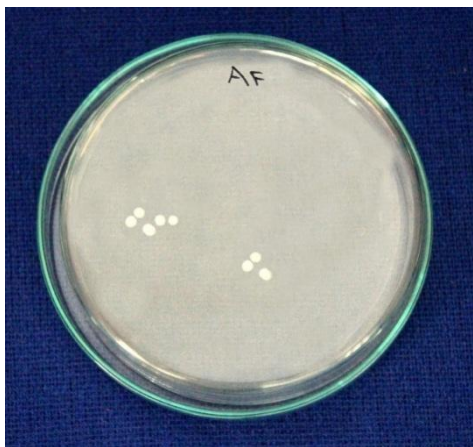
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**Figure 3: Microbial**

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**Plate View Day 1**

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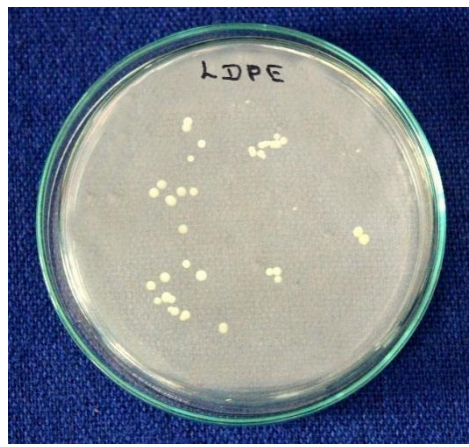


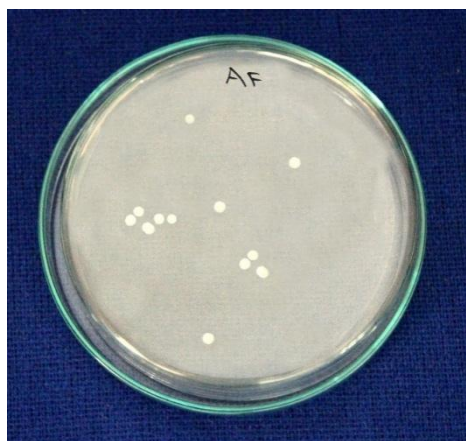
Figure 4: Microbial Plate View Day 15

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Figure 5: Microbial Plate View Day 30



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272 Table 5: Organisms Identified for Cookies

Day	Samples	Organisms Identified
Day-1 <sup>st</sup>	AF	<i>Lactobacillus bulgaricus</i>
	LDPE	<i>Lactobacillus lactis</i>
	HDPE	<i>Streptococcus thermophilus</i>
Day-15 <sup>th</sup>	AF	<i>Lactobacillus casei</i>
	LDPE	<i>Pseudomonas mephitica</i>
	HDPE	<i>Pseudomonas nigrificans</i>
Day-30 <sup>th</sup>	AF	<i>Lactobacillus acidophilus</i>

	LDPE	<i>Bifidobacterium longum</i>
	HDPE	<i>Bacillus polymyxa</i>

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274 The presence of these organisms during the fermentation periods confirms that they grow in  
 275 close association with the food substrate and produce extracellular enzymes [32].

276

## 277 **Conclusion**

278 In recent years, medicinal plant conservation and use have received a lot of attention.  
 279 It has been utilised by indigenous and marginal people around the world to treat a variety of  
 280 ailments from time immemorial. The goal of this study was to assess the nutritional value,  
 281 mineral content, and microbial activity of healthy cookies. When compared to other  
 282 concentrations and controls, the 0.50 percent concentration of *G.sylvestre* demonstrated  
 283 superior activity. *G.sylvestre* was a traditional medicinal plant whose leaves had remarkable  
 284 therapeutic characteristics and were highly efficient for diabetic therapy. As a result,  
 285 *Gymnema* leaves can be utilised in the preparation of hypoglycemic mix-in foods. According  
 286 to the findings of the study, these meal preparations may aid in instilling the habit of taking  
 287 herbal mixtures, which are curative and therapeutic for diabetes patients. As a result, this  
 288 study showed that we must raise knowledge about the usage of locally available but  
 289 underutilised medicinal herbs.

## 290 **Ethical Approval:**

291 As per international standard or university standard ethical approval has been collected and  
 292 preserved by the authors.

293

## 294 **NOTE:**

295 The study highlights the efficacy of " Ayurvedic component " which is an ancient  
 296 tradition, used in some parts of India. This ancient concept should be carefully evaluated in  
 297 the light of modern medical science and can be utilized partially if found suitable

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