IJCRT.ORG ISSN: 2320-2882



INTERNATIONAL JOURNAL OF CREATIVE RESEARCH THOUGHTS (IJCRT)

An International Open Access, Peer-reviewed, Refereed Journal

Development And Evaluation Of Fermented Bottle Gourd Beverages Using Various Sugar Sources: Biochemical, Functional And Sensory Analysis

¹Dr. Naomita Dhume*, ²Ms. Mariyah Patel*, ³Ms. Misba Mustak Patel*, ⁴Ms. Palvi Satish Kamat, ⁵Mrs. Maria Seelan Nadar

¹Head of the Department, ²Lecturer, ³Student, ⁴Assistant Professor, ⁵Assistant Professor

Department of M.Sc. Bioanalytical Sciences

Guru Nanak Khalsa College o<mark>f Arts, Science and Commerce (Autonomous); Nathalal Parekh Marg, Matunga (East), Mumbai 400 019, Maharashtra, India</mark>

Abstract: Lagenaria siceraria (bottle gourd), a Cucurbitaceae vine cultivated across tropical regions, is rich in moisture, fiber, vitamins, phytosterols, flavonoids, terpenoids, cucurbitacins, and bioactive polysaccharides. Its traditional uses—across fruit, leaves, seeds, and flowers—include managing diabetes, hypertension, liver disorders, insomnia, urinary issues, constipation, pain, and inflammation. Modern studies demonstrate antioxidant, antihyperlipidemic, antihyperglycemic, cardioprotective, hepatoprotective, immunomodulatory, diuretic, analgesic, anti-inflammatory, anthelmintic, adaptogenic, anticancer, and neuroprotective effects. Its broad pharmacological profile supports its role as a functional nutraceutical and medicinal food, particularly in metabolic and chronic disease prevention. Fermented bottle gourd beverages were prepared from 4 types of sugars (Table Sugar, Rock Sugar, Sugarcane Jaggery and Palm Jaggery). The study aimed to evaluate the influence of these sugar sources on the biochemical, functional, and sensory properties of fermented bottle gourd beverages. Fermentation significantly reduced pH, enhanced stability, and improved the release of phenolic compounds. Among the formulations, the palm jaggery-based beverage exhibited the highest antioxidant activity, maximum total phenolic content, elevated reducing sugars and the greatest ethanol yield, demonstrating superior functional enhancement during fermentation. Sugarcane jaggery also produced nutritionally favorable beverages, whereas refined sugars showed comparatively lower functional attributes. Sensory evaluation revealed strong consumer acceptability for all sugar-based formulations, with palm jaggery scoring highest for its distinctive aroma, color, and balanced taste profile. Overall, the findings highlight palm jaggery as the most effective sugar source for producing a nutritionally enriched, functionally potent, and sensorially appealing fermented bottle gourd beverage. The study underscores the potential of integrating traditional sweeteners with fermentation to develop affordable, value-added nutraceutical beverages with promising applications in the functional food and health industry.

Index Terms: Bottle gourd, beverages, cucurbitacins, sensory evaluation, ethanol, antioxidant activity

I. INTRODUCTION:

Bottle gourd belongs to the Cucurbitaceae family (cucumber, squash, etc.) with the scientific name Lagenaria siceraria (Mol.) [1]. This family has around 130 genera and 800 species [2]. Bottle gourd is also known as Calabash, Doodhi, and Lauki in different parts of India. It is yellowish green, having the shape of a bottle with white pulp. India, Sri Lanka, South Africa, Indonesia and Malaysia are the major bottle gourd producing countries in the world [1]. The plant can grow on the ground like a pumpkin vine, and it is a herbaceous plant with branches in nature of their growth. Their stems are covered with short soft hair known as downy. The leaves are alternately arranged, and tendrils are also consistently found. The roots are smooth and circular in shape and white or pale cream. The taproot grows up to a distance of 80cm downward, but most of the roots spread out and occupy the upper soil. Bottle gourd has both male and female flowers known as monoclines found on different sections of a vine; because of that, higher cross-pollination occurs. There are various types of bottle gourd shapes that belong to the different kind of genre having various shapes and sized fruit, narrow neck, elongated, and round. The seeds are flat, approximately rectangular, or trapezoidal, whitish to dark brown at the ends. Generally, fruit length depends on its variety and varies from 150 to 1000mm. The overall ingredients of the fruit are essential for the better and healthier lives of human beings [3]. Bottle gourd is one of the excellent fruits gifted by the nature to human beings having composition of all the essential constituents that are required for normal and good human health [1]. It approximately contains moisture 94.5±0.06 %, protein 1.2±0.06 %, fat 0.2±0.02 %, carbohydrate 3.75±0.03 %, fibre 0.7 ± 0.01 %, ash 0.5 ± 0.01 %, and energy 15 ± 0.12 % [1,3].

Table 1: Taxonomical classification of Bottle gourd [4]

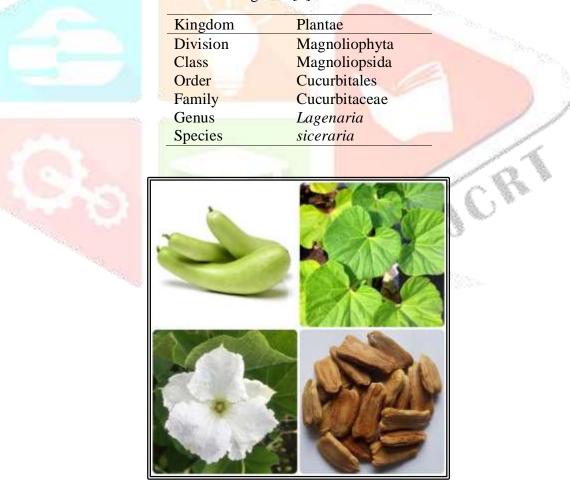


Fig 1: Different parts of Bottle gourd plant [5, 6, 7, 8]

Bottle gourd has long been an important component of indigenous herbal medicines, particularly in Asia. Its leaves, seeds and flowers also have medicinal applications. Its consumption is advocated by traditional healers for controlling diabetes mellitus, hypertension, liver diseases, weight loss and other associated benefits. It is well known that bottle gourd is helpful in constipation, premature graying of hair, urinary disorders and insomnia which reflect significant health-promoting properties [1].

The phytochemical analysis of edible portion of the fruit it is shown that it is a good source glucose and fructose. The fruit is a good source of vitamins B and a fair source of ascorbic acid [4]. Bottle gourd also contains terpenoids, which are a class of natural compounds found in plants. These terpenoids contribute to the plant's medicinal properties and various pharmacological activities [9, 10]. Specifically, bottle gourd contains triterpenoids, including cucurbitacins which are known for their diverse biological activities [4, 9, 10]. Cucurbitacins are bitter compounds that acts as a defence mechanism against insects and other herbivores. Studies have shown that cucurbitacin has a promising pharmacological properties but increased levels lead to toxicity, capillary leak, hypotension, polyneuritis. There are four subtypes of cucurbitacins which are B, D, G & H. The Cucurbitacin D is most potent and has shown increase capillary permeability which in turn reduces blood pressure, causes ascites and pleural effusion. Toxicity in humans is primarily due to this compound [11]. Leaves contain cucurbitacin B and roots, cucurbitacins B, D and E [4]. Other terpenoids, like fucosterol and campesterol, have also been identified. These compounds are found in different parts of the plant, including the fruit, seeds, and leaves [4, 9, 10].

1.1 Pharmacological properties of Bottle gourd

Lagenaria siceraria possesses excellent pharmacological properties that can be used to treat different diseases. Some of the properties are elaborated below:

- 1.1.1 Anti-hyperlipidemic property- The administration of the aqueous extract of bottle gourd orally reduced the elevated levels of triglycerides, cholesterol, and low-density lipoprotein while enhancing the high-density lipoprotein levels. The dietary fibre present in the fruit lowers the cholesterol level. Saponins in this fruit increase lipoprotein activity and rapidly remove fatty acids in the blood [3].
- 1.1.2 Anti-inflammatory and analgesic characteristics- Ghule and co-workers showed the anti-inflammatory and analgesic effects of bottle gourd juice extract in mice and rats. They studied its analgesic property on acetic acid-induced writhing and formalin pain tests on mice. The anti-inflammatory effects were investigated using acute inflammatory models, i.e., ethyl phenyl propionate induced ear edema, carrageenan, arachidonic acid induced hind paw edema, and the albumin-induced paw edema in rats. The activity was due to the presence of flavonoids [3].
- 1.1.3 <u>Diuretic property</u>- Diuretic activity of *Lagenaria siceraria* fruit was assessed by measuring different parameters, i.e., total urine volume, urine concentration of sodium, potassium, and chloride. It was found that the extracts of *Lagenaria siceraria* fruit (100-200mg kg-1, p.o.) showed higher urine volume and exhibited dose-dependent increase in excretion of electrolytes [3].
- 1.1.4 Antioxidant property- Acetone extract of fruit epicarp of *Lagenaria siceraria* fruit showed maximum antioxidant activity in-vitro model using DPPH. The fresh juice of the fruit also showed free radical scavenging activity. The fruit extract was also effective in CCl₄ induced liver damage, where it maintained the level of endogenous antioxidant enzymes (superoxide dismutase, catalase and glutathione peroxidase) and marker of lipid peroxidation to normal [3].
- 1.1.5 <u>Immunomodulatory property</u>- When extracted with methanol, bottle gourd fruit showed notable reactions in rats, increasing white cells and lymphocytes. HPLC analysis showed the presence of lagenin (a ribosome-inactivating protein), which is responsible for the activity. Bottle gourd was evaluated for its immunomodulatory activity against pyrogallol induced immunosuppression. The results showed an increase in humoral and cellular immunity. The increase in non-specific immunity is indicated by the subsequent rise in the total leucocyte and neutrophils count. The studies made it clear that the fruit has both specific and non-specific immunity modulating activity [3].
- 1.1.6 <u>Hepato and cardioprotective property</u>- The fruit powder of *Lagenaria siceraria* showed good cardioprotective effects. The drug was studied against Doxorubicin induced cardiotoxicity in rats at 200mg/kg, p.o. for 18 days. The *Lagenaria siceraria* prevented the alteration in endogenous antioxidants (superoxide dismutase, reduced glutathione) and lipid peroxidation, whereas markers of cardiotoxicity i.e., CK, MB, and LDH were significantly reduced. Further, the

Lagenaria siceraria powder also showed protection against changes in ECG and histopathological alterations induced by doxorubicin. Ethanolic extract of Lagenaria siceraria fruits also showed the increased force of contraction and decreased rate of contraction (from 66 to 44) in isolated frog hearts, when perfused with normal ringer solution [3].

1.1.7 <u>Anthelmintic property</u>- The ethanolic extracts of the seeds of *Cucumis sativus*, *Cucurbita maxima* and *Lagenaria siceraria* exhibited potent anthelmintic activity against tapeworms, which was comparable to the effect of piperazine citrate [3].

Bottle gourd takes the lead in the diet compared to other vegetables grown in India since it is a nutrient-dense and low-cost source. Non-bitter bottle gourd is used for eating. The various products that can be developed from bottle gourd fruit are pickles, chutney, juice, sweets (barfi, halwa), candy and bakery and dairy products (kheer, pedha, biscuits) [3]. One such product which can be produced from Bottle gourd is a fermented beverage.

Fermented beverages can also be produced using bottle gourd but it is not demonstrated widely. Hence, detailed technique of such beverage and its properties is insufficient. Bottle gourd does not have ample sugar level to undergo the alcoholic fermentation; therefore, sugar is required to be added during fermented bottle gourd beverage making as compared to classical fermented beverages making. In this research, fermented bottle gourd beverages are produced which are unique products made from bottle gourd fruit using different sources of sugars; viz. Table Sugar, Rock Sugar, Sugarcane Jaggery, Palm Jaggery. A control fermented beverage (without addition of any sugar) is also prepared.

Initially, bottle gourd juice extracted from bottle gourd of a local market of Maharashtra is analysed to check for its anti-oxidant activity, protein content, reducing sugar content, mineral content. Then fermented beverages are prepared of the same bottle gourd fruit.

Sensory evaluation and qualitative phytochemical profiling (flavonoids, triterpenoids) of fermented bottle gourd beverages is performed and they are examined for their pH, miscibility, purity, ethanol content, anti-oxidant activity, protein content, reducing sugar content, mineral content, total phenolic content estimation.

- II. MATERIALS AND METHODS:
- 2.1 Bottle gourd juice extraction and analysis:
- 2.1.1 Bottle gourd juice extraction-

Fresh bottle gourd was procured locally in Maharashtra and checked for its authenticity. After authentication, the bottle gourd was thoroughly washed in running water to remove adhering foreign particles, both the ends were cut and disposed. The edible portion was cut into small pieces along with the peel and blended. After blending, the extracted juice was filtered through muslin cloth. Before extraction of the juice, the grinder's jar was thoroughly washed to ensure no impurities gets mixed with the juice. The extracted juice was collected in a clean beaker for analysis.

I CR

- 2.1.2 Bottle gourd juice analysis-
- **2.1.2.1 pH-** pH was measured using a digital pH meter (Model: LABMAN LMPH-9).
- **2.1.2.2 Antioxidant Assay (DPPH Assay)-** Following [12, 13] with tailored adjustments, assays were performed using 0.4 mL juice (or 0.4 mL methanol for the blank), plus 4.0 mL of 0.1 mM methanolic DPPH and 1.6 mL of 0.1 M Tris-HCl. Mixtures were vortexed, incubated in the dark for 30 min, and absorbance read at 520 nm using UV-Vis Spectrophotometer (Model: Shimadzu UV-1650PC). %RSA was then calculated.

 $\%RSA = \frac{Absorbance\ of\ blank-Absorbance\ of\ sample}{Absorbance\ of\ blank} \times 100$

- 2.1.2.3 Protein Estimation- A modified protocol based on [14, 15] was employed. Standard BSA solutions (100–500 ppm) were prepared by mixing 0.1–0.5 mL of 1000 ppm BSA stock with 0.9–0.5 mL distilled water. A fresh 1:1 Alkaline CuSO₄–Folin-Ciocalteu reagent was prepared. For the blank, 1.0 mL distilled water was used. Juice sample was diluted 1:5 (juice:water), and 1.0 mL of each dilution was mixed with 5.0 mL Alkaline CuSO₄, vortexed, and incubated for 10 min. Then, 0.5 mL Folin-Ciocalteu reagent was added, followed by vortexing and a 30 min room-temperature incubation. Absorbance was measured at 660 nm using UV-Vis Spectrophotometer (Model: Shimadzu UV-1650PC). Protein content (ppm) was calculated from the standard curve (absorbance vs. concentration).
- 2.1.2.4 Reducing Sugar Estimation- A modified protocol based on [16, 17] was applied. Standards (100–500 ppm glucose) were prepared by adding 0.2–1.0 mL of 1000 ppm glucose stock to labeled tubes, topped up with distilled water (total volume ~2.0 mL). DNSA reagent was freshly prepared. For the blank, 2.0 mL distilled water was used. Juice sample was diluted 1:40 (juice:water), and 2.0 mL of each dilution was mixed with 2.0 mL DNSA reagent, vortexed, covered with aluminum foil, and heated in a boiling water bath for 5 min. After cooling, absorbance was measured at 540 nm using UV-Vis Spectrophotometer (Model: Shimadzu UV-1650PC). Reducing sugar concentration (ppm) was determined by interpolation from the standard curve (absorbance vs. concentration).
- 2.1.2.5 Mineral Estimation- An adapted method from [18] was used to estimate Na⁺ and K⁺ via flame photometry (Model: EQ 850A). Stock solutions of 1000 ppm NaCl and KCl were prepared and serially diluted to 20 ppm via intermediate standards (1000 → 100 → 50 → 20 ppm). Further working standards (1, 5, 10, 15, 20 ppm) were prepared from the 50 ppm solution. Juice samples were diluted (Na⁺: 1:4; K⁺: 1:49 juice:water). Distilled water was aspirated to set a zero baseline. The highest standard (20 ppm) was aspirated next, and the instrument sensitivity was adjusted to full-scale (galvanometer = 100). Subsequent standards (1–15 ppm) were aspirated in ascending order, with readings recorded. Samples were analyzed the same way, interspersed with distilled water rinses. Na⁺ and K⁺ concentrations (ppm) were then determined via calibration from the standard curve.

2.2 Raw Materials (Sugars) Analysis:

Antioxidant Assay (DPPH Assay)- For antioxidant assay of sugars, the procedure followed was the same as described in [12, 13] and performed for bottle gourd juice with modifications. 0.5 mM Methanolic DPPH was prepared instead of 0.1 mM solution. Sample solutions were prepared by dissolving 2 g of each sugars in 5 ml of distilled water. Those sample solutions were added in a volume of 0.4 ml in respective labelled test tubes instead of juice and the remaining analyses was subsequently conducted.

2.3 Fermented bottle gourd beverages preparation and analysis:

2.3.1 Fermented bottle gourd beverages preparation-

After performing the tests for bottle gourd juice, the bottle gourd fruit was utilised for fermented beverage production.

The bottle gourd weighing approx. 1 kg was thoroughly washed and cut into pieces. The gourd (along with the seeds and rind) was blended in a mixer grinder. After blending, the extracted juice was separated from the pulp using a muslin cloth. Whilst blending, 4 jars were submerged in hot water for 20-30 mins for sterilisation. 250 ml of juice was measured and poured in each jar, together with 250 ml of potable water (amount of bottle gourd juice = amount of water) [19]. The jars were labelled as Ferme. BG Table Sugar Bev., Ferme. BG Rock Sugar Bev., Ferme. BG Sugarcane Jaggery Bev., Ferme. BG Palm Jaggery Bev. (BG means Bottle

Gourd, Ferme. means Fermented, Bev. means Beverage). The 4 different sugars; Table Sugar, Rock Sugar, Sugarcane Jaggery, Palm Jaggery; all weighing 100 g were added to their respective labelled jars. Then 12.5 g activated Baker's yeast was added to each jar. All the ingredients were mixed properly. Since, yeast converts sugars into alcohol and carbon dioxide. So, to release the excess carbon dioxide which was coming out of the closed jars in the form of foam, they were kept a bit opened under sterilised environment. Once, the sugars were completely dissolved and excess carbon dioxide was released, the jars were sealed making sure no air enters into the jars. The beverages were allowed to ferment under dark conditions. After few days, a fermented control beverage was also prepared. It was prepared the same way as other beverages were, the only difference was, no sugar was added to the juice. Every week for 1 year, the beverage jars were shaken so that the yeast gets used up. The drinks were then filtered and transferred to other sterilised jars according to their labels. Sensory evaluation and qualitative phytochemical profiling (flavonoids, triterpenoids) of the beverages was performed and were also examined for their pH, miscibility, purity, ethanol content, anti-oxidant activity, protein content, reducing sugar content, mineral content and total phenol content.



Fig 2: Flow Chart of Fermented bottle gourd beverages preparation

2.3.2 Fermented bottle gourd beverages analysis-

2.3.2.1 Qualitative Phytochemical Profiling:

Fermented bottle gourd beverages were analysed for their Phytochemical characteristics as per standard methods given in [20, 21, 22].

Table 2: Qualitative Phytochemical Profiling of beverages

Sr. No. Tests Tests description 2.3.2.1.1 Test for Flavonoids FeCl ₃ Test- [20] 2 ml of each beverage was mixed with few drops of FeCl ₃ separately in test tubes. The solutions were observed for their colour change to green. Shinoda Test- [21] 2 ml of each beverage was mixed with 1 ml conc. HCl and a pinch of Mg ribbon. The solutions were observed for their colour change to pink/ red/ orange. 2.3.2.1.2 Test for Triterpenoids Salkowski's Test- [22] 2 ml of beverages were separately mixed with few drops of conc. H ₂ SO ₄ . The solutions were observed for their lower layer turning to yellow colour. Liebermann Burchard Test- [22] 2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were observed for colour change to eosin red.	<u>.</u>	Table 2: Q	Quantative Phytochemica	i Proming of beverages
2 ml of each beverage was mixed with few drops of FeCl ₃ separately in test tubes. The solutions were observed for their colour change to green. Shinoda Test- [21] 2 ml of each beverage was mixed with 1 ml conc. HCl and a pinch of Mg ribbon. The solutions were observed for their colour change to pink/ red/ orange. Salkowski's Test- [22] 2 ml of beverages were separately mixed with few drops of conc. H ₂ SO ₄ . The solutions were observed for their lower layer turning to yellow colour. Liebermann Burchard Test- [22] 2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were		Sr. No.	Tests	Tests description
few drops of FeCl ₃ separately in test tubes. The solutions were observed for their colour change to green. Shinoda Test- [21] 2 ml of each beverage was mixed with 1 ml conc. HCl and a pinch of Mg ribbon. The solutions were observed for their colour change to pink/ red/ orange. 2.3.2.1.2 Test for Triterpenoids Salkowski's Test- [22] 2 ml of beverages were separately mixed with few drops of conc. H ₂ SO ₄ . The solutions were observed for their lower layer turning to yellow colour. Liebermann Burchard Test- [22] 2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were		2.3.2.1.1	Test for Flavonoids	FeCl ₃ Test- [20]
few drops of FeCl ₃ separately in test tubes. The solutions were observed for their colour change to green. Shinoda Test- [21] 2 ml of each beverage was mixed with 1 ml conc. HCl and a pinch of Mg ribbon. The solutions were observed for their colour change to pink/ red/ orange. 2.3.2.1.2 Test for Triterpenoids Salkowski's Test- [22] 2 ml of beverages were separately mixed with few drops of conc. H ₂ SO ₄ . The solutions were observed for their lower layer turning to yellow colour. Liebermann Burchard Test- [22] 2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				2 ml of each beverage was mixed with
tubes. The solutions were observed for their colour change to green. Shinoda Test- [21] 2 ml of each beverage was mixed with 1 ml conc. HCl and a pinch of Mg ribbon. The solutions were observed for their colour change to pink/ red/ orange. Salkowski's Test- [22] 2 ml of beverages were separately mixed with few drops of conc. H ₂ SO ₄ . The solutions were observed for their lower layer turning to yellow colour. Liebermann Burchard Test- [22] 2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				few drops of FeCl ₃ separately in test
their colour change to green. Shinoda Test- [21] 2 ml of each beverage was mixed with 1 ml conc. HCl and a pinch of Mg ribbon. The solutions were observed for their colour change to pink/ red/ orange. 2.3.2.1.2 Test for Triterpenoids Salkowski's Test- [22] 2 ml of beverages were separately mixed with few drops of conc. H ₂ SO ₄ . The solutions were observed for their lower layer turning to yellow colour. Liebermann Burchard Test- [22] 2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				· · · · · · · · · · · · · · · · · · ·
Shinoda Test- [21] 2 ml of each beverage was mixed with 1 ml conc. HCl and a pinch of Mg ribbon. The solutions were observed for their colour change to pink/ red/ orange. 2.3.2.1.2 Test for Triterpenoids Salkowski's Test- [22] 2 ml of beverages were separately mixed with few drops of conc. H ₂ SO ₄ . The solutions were observed for their lower layer turning to yellow colour. Liebermann Burchard Test- [22] 2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				
2.3.2.1.2 Test for Triterpenoids Salkowski's Test- [22] 2 ml of beverages were separately mixed with few drops of conc. H ₂ SO ₄ . The solutions were observed for their lower layer turning to yellow colour. Liebermann Burchard Test- [22] 2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				men corour enunge to green.
ml conc. HCl and a pinch of Mg ribbon. The solutions were observed for their colour change to pink/ red/ orange. 2.3.2.1.2 Test for Triterpenoids Salkowski's Test- [22] 2 ml of beverages were separately mixed with few drops of conc. H ₂ SO ₄ . The solutions were observed for their lower layer turning to yellow colour. Liebermann Burchard Test- [22] 2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				
The solutions were observed for their colour change to pink/ red/ orange. 2.3.2.1.2 Test for Triterpenoids Salkowski's Test- [22] 2 ml of beverages were separately mixed with few drops of conc. H ₂ SO ₄ . The solutions were observed for their lower layer turning to yellow colour. Liebermann Burchard Test- [22] 2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				
colour change to pink/ red/ orange. 2.3.2.1.2 Test for Triterpenoids Salkowski's Test- [22] 2 ml of beverages were separately mixed with few drops of conc. H ₂ SO ₄ . The solutions were observed for their lower layer turning to yellow colour. Liebermann Burchard Test- [22] 2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				ml conc. HCl and a pinch of Mg ribbon.
2.3.2.1.2 Test for Triterpenoids Salkowski's Test- [22] 2 ml of beverages were separately mixed with few drops of conc. H ₂ SO ₄ . The solutions were observed for their lower layer turning to yellow colour. Liebermann Burchard Test- [22] 2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				The solutions were observed for their
2 ml of beverages were separately mixed with few drops of conc. H ₂ SO ₄ . The solutions were observed for their lower layer turning to yellow colour. Liebermann Burchard Test- [22] 2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				colour change to pink/ red/ orange.
2 ml of beverages were separately mixed with few drops of conc. H ₂ SO ₄ . The solutions were observed for their lower layer turning to yellow colour. Liebermann Burchard Test- [22] 2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were	-	ATA ATA	Total View	
with few drops of conc. H ₂ SO ₄ . The solutions were observed for their lower layer turning to yellow colour. **Liebermann Burchard Test-[22]* 2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. **Tschugajen's Test-[20]* 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were		2.3.2.1.2	Test for Triterpenoids	200
Liebermann Burchard Test- [22] 2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				
Liebermann Burchard Test- [22] 2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were	-0000			with few drops of conc. H ₂ SO ₄ . The
Liebermann Burchard Test- [22] 2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				solutions were observed for their lower
2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				layer turning to yellow colour.
2 ml of beverages were mixed with few drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				
drops of acetic acid and 1 ml conc. H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				Liebermann Burchard Test- [22]
H ₂ SO ₄ . The solutions were observed for deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				2 ml of beverages were mixed with few
deep red colour at the junction of the tubes. Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				drops of acetic acid and 1 ml conc.
Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				H ₂ SO ₄ . The solutions were observed for
Tschugajen's Test- [20] 2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				deep red colour at the junction of the
2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				
2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were				
2 ml of beverages were mixed with 5 ml of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were	The same of			Tschugajen's Test-[20]
of acetyl chloride and pinch of ZnCl ₂ . The solutions were warmed in a hot water bath for few minutes. The solutions were	100			7.1
solutions were warmed in a hot water bath for few minutes. The solutions were		4.0	1	
		N 196		TOP W
			All St.	
33321.33 131 331332 311113 10 3 3111131				
				6

2.3.2.2 Physicochemical analysis:

2.3.2.2.1 pH- pH of fermented bottle gourd beverages was determined using a calibrated digital pH digital pH meter (Model: LABMAN LMPH-9) [23].

2.3.2.2.2 Stability Test-

- a) Colour Stability- The colour stability assessment of the fermented bottle gourd beverages was performed following the standardized methodology described by Enartis
- b) Protein / Heat Stability- This stability test of the fermented beverages was administered as per the methodology given by Iowa State University [25].
- c) Cold Stability- Cold Stability testing of the fermented beverages was carried out in accordance with the protocol given by Australian Wine Research Institute [26].

- **2.3.2.2.3 Antioxidant Assay-** For antioxidant assay of beverages, the procedure followed was the same as described in [12, 13] and performed for bottle gourd juice and sugars with modifications. 0.5 mM Methanolic DPPH was prepared instead of 0.1 mM of the solution and the remaining experimental procedure was executed accordingly.
- **2.3.2.2.4 Protein Estimation** Beverages' protein estimation was conducted as per the methodologies detailed in [14, 15], adjusted specifically for bottle gourd juice and subsequently conducted.
- **2.3.2.2.5 Reducing Sugar Estimation-** Reducing sugar estimation of beverages was carried out according to the procedures described in [16, 17], incorporating specific modifications for use with bottle gourd juice and subsequent procedure was carried out to complete the analysis.
- **2.3.2.2.6 Total Phenolic Content Estimation-** The total phenolic content was determined using the Folin–Ciocalteu method as demonstrated by Miranda J. [27] and instrument used was UV-Vis Spectrophotometer (Model: Shimadzu UV-1650PC).
- **2.3.2.2.7 Mineral Estimation** (Na⁺ and K⁺)- The beverages' mineral estimation protocol was also adopted from [18] and procedure followed was the same as customised for bottle gourd juice.
- 2.3.2.2.8 Purity of Volatile Organic Compounds- Initially, the fermented bottle gourd beverages were filtered using 0.45 μm filter paper and outsourced to check the content of the volatile organic compounds, i.e., Ethanol, dimethyl ether, 1-methoxy-2-methylbutane, 1-chloropentane, 3-methyl-2-pentanol, 2,3-butanediol, glycerin, N-methoxy-methanamine were analysed. Out of which Purity of Ethanol in the beverages was found and reported.
- 2.3.2.2.9 Ethanol Content Estimation- The procedure and preparation of test solutions were carried out according to a formerly published procedure [28] with slight modifications. Ethanol standards were prepared from 99.5% ethanol in the range of 10% to 50% v/v standard calibration curve. The test solutions were prepared by adding 640 μl of sample or standard, 2560 μl of 0.05 M K₂Cr₂O₇. Mixing and allowing the solution to stand for 10 mins, then adding 140 μl of conc. H₂SO₄ to those solutions. Measurements of absorbance were performed with UV-Vis Spectrophotometer (Model: Shimadzu UV-1650PC) at 535 nm. % v/v of ethanol in beverages were found out through standard calibration curve.

2.3.2.3 Sensory evaluation:

The sensory evaluation of all five fermented bottle gourd beverages was being conducted by semi-trained panel of judges by using composite scoring and hedonic rating test. Coded samples were given to 12 judges. They were asked to rinse their mouth before or in between testing the given sample. Each sample was evaluated for overall acceptability on hedonic scale (5 point) and composite scores using various quality attributes viz. colour and appearance, odour (fruity, spicy, vanilla, woody), taste (sour, bitter, salty, sweet), mouthfeel (astringent, alcohol, body) and overall impression on a stipulated pro forma [29, 30].

III. RESULTS:

3.1 Results of bottle gourd juice analysis:

Table 3: Results of bottle gourd juice analysis

Juice	pН	%RSA (%)	Protein (ppm)	Reducing	Minerals	
				sugars (ppm)	Na ⁺ (ppm)	K ⁺ (ppm)
Bottle gourd Juice	5.82	80.70	860.134	601.385	40.165	1030

3.2 Result of Antioxidant Assay (DPPH Assay) of raw materials (sugars):

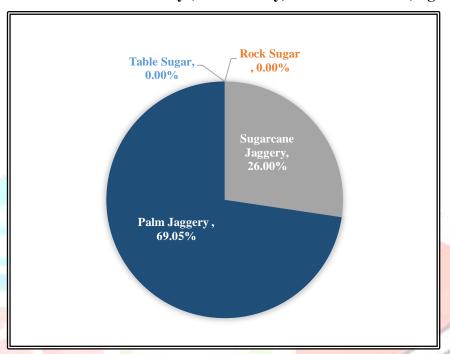


Fig 3: Pie chart of % RSA (%) of beverages

3.3 Results of fermented bottle gourd beverages analysis:

3.3.1 Qualitative Phytochemical Profiling:

Table 4: Results of Qualitative Phytochemical Profiling of beverages

Sr. No.	Fermented bottle	Test for Flavonoids		Tests for Triterpenoids		
	gourd beverage	FeCl ₃ Test	Shinoda Test	Salkowski Test	Liebermann Burchard Test	Tschugajen's Test
1.	Ferme. BG Table	+	-	-	-	-
	Sugar Bev.					
2.	Ferme. BG Rock	+	-	-	-	-
	Sugar Bev.					
3.	Ferme. BG	+	-	-	-	-
	Sugarcane					
	Jaggery Bev.					
4.	Ferme. BG Palm	-	-	-	-	-
	Jaggery Bev.					
5.	Ferme. BG	-	-	-	-	-
	Control Bev.					

3.3.2 Physicochemical Analysis:

3.3.2.1 pH-

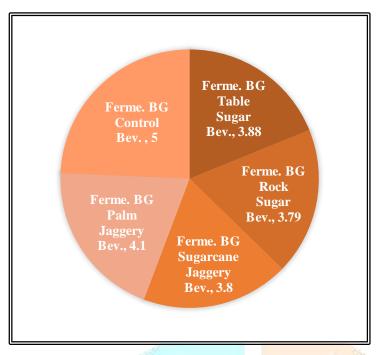


Fig 4: Pie chart of pH of beverages

3.3.2.2 Stability Test-

Table 5: Results of Stability Test

Sr. No.	Fermented bottle gourd beverage	Stability					
1.	Ferme. BG Table	Stable					
	Sugar Bev.	Sugar Bev.					
2.	Ferme. BG Rock	Stable					
	Sugar Bev.						
3.	Ferme. BG	Stable					
	Sugarcane Jaggery						
	Bev.						
4.	Ferme. BG Palm	Stable					
	Jaggery Bev.						
5.	Ferme. BG Control	Stable					
	Bev.						

3.3.2.3 Antioxidant Assay-

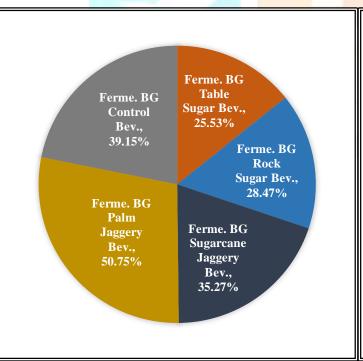


Fig 5: Pie chart of % RSA (%) of protein (ppm) in beverages

3.3.2.4 Protein Estimation-

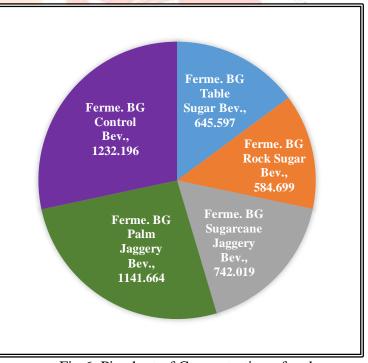
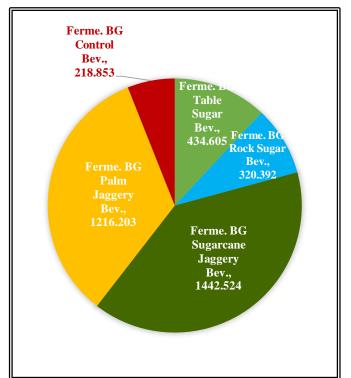


Fig 6: Pie chart of Concentration of beverages

3.3.2.5 Reducing Sugar Estimation-

3.3.2.6 Total Phenolic Content

Estimation-



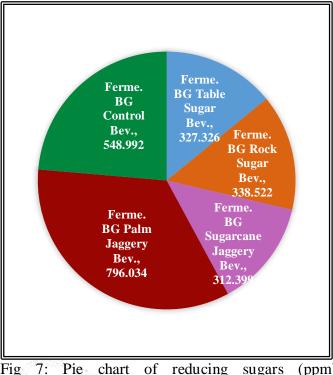


Fig 8: Pie chart of total phenols (ppm) beverages

Fig 7: Pie chart of reducing sugars (ppm) in beverages in

3.3.2.7 Mineral Estimation (Na⁺ and K⁺)-

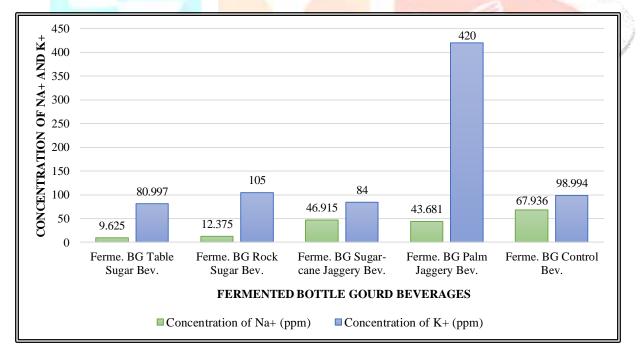


Fig 9: Bar graph of Concentration of Minerals (Na⁺ and K⁺) (ppm) in beverages

3.3.2.8 Ethanol Content Estimation-(Ethanol)-

3.3.2.9 Purity of Volatile Organic Compound

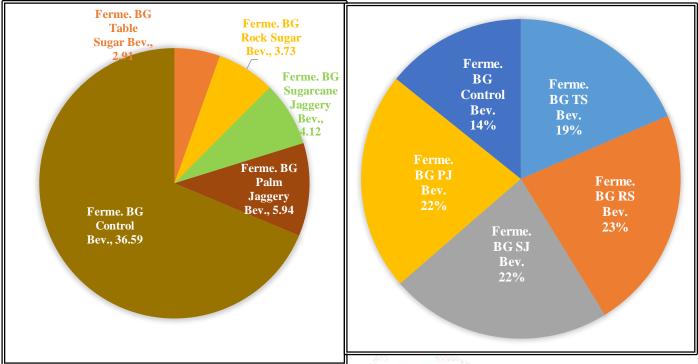


Fig 11: Pie chart of purity of ethanol in beverages beverages

Fig 10: Pie chart of ethanol content (% v/v) (%) in

3.3.3 Sensory Evaluation:

Table 6: Results of Sensory Evaluation of beverages

						Contract of the Contract of th
Sr. No.	Fermented bottle gourd beverage	Colour	Odour	Taste	Mouthfeel	Overall acceptability (Hedonic scale)
1.	Ferme. BG	Caramel	Fruity	Sour	Astringent	5
	Table Sugar	Brown				
	Bev.	1000		J		
2.	Ferme. BG	Caramel	Fruity	Sour	Astringent	5
	Rock Sugar	Brown				
	Bev.					
3.	Ferme. BG	Caramel	Fruity	Sour	Astringent	5
	Sugarcane	Brown				
	Jaggery Bev.					
4.	Ferme. BG	Black	Vanilla	Sour	Body	5
	Palm Jaggery					
	Bev.					
5.	Ferme. BG	Beige	Pungent	Bitter	Body	1
	Control Bev.					

(Hedonic Scale: 1- Dislike, 2- Neither like, 3- Like slightly, 4- Like moderately, 5- Like very much)

IV. DISCUSSION:

The present study aimed to develop and evaluate fermented bottle gourd beverages using different sugar sources, namely—table sugar, rock sugar, sugarcane jaggery, and palm jaggery—and to assess their biochemical, functional, and sensory characteristics. The findings clearly demonstrate that the type of sugar used during fermentation significantly influenced the beverage's antioxidant activity, nutrient composition, ethanol yield, and overall acceptability.

4.1 Effect of Fermentation on pH and Stability-

Fermentation of bottle gourd juice led to a marked reduction in pH across all sugar-containing formulations, ranging from 3.79 to 4.1, compared to the control beverage (pH 5.0). This acidification can be attributed to yeast metabolism, which produces organic acids and ethanol during sugar breakdown. The lower pH not only supports microbial stability but also enhances product preservation. All beverages exhibited satisfactory physical and thermal stability, confirming that the fermentation process yielded shelf-stable beverages without phase separation or spoilage.

4.2 Antioxidant and Phenolic Profile-

A significant variation in antioxidant potential was observed among beverages, with the palm jaggery-based sample showing the highest radical scavenging activity (50.75 % RSA), followed by sugarcane jaggery (35.27 % RSA). These results align with the intrinsic antioxidant properties of jaggery, which contains polyphenols, minerals, and vitamins absent in refined sugars. Fermentation is also known to liberate bound phenolic compounds, thereby enhancing antioxidant capacity. The total phenolic content further confirmed this trend, where palm jaggery beverages exhibited the highest concentration (796.03 ppm). Such enhancement underscores the synergistic effect between the natural antioxidants of jaggery and fermentation-induced biochemical transformations.

4.3 Protein and Reducing Sugar Content:

Protein content decreased moderately after fermentation compared to unfermented juice, as proteins serve as nitrogen sources for yeast growth. However, the control beverage showed slightly higher residual protein (1232.19 ppm), possibly due to limited fermentation. The reducing sugar content was markedly higher in jaggery-based beverages, indicating that complex carbohydrates in jaggery undergo partial hydrolysis during fermentation. Palm jaggery again yielded the highest reducing sugar value (1141.66 ppm), suggesting incomplete sugar utilization or slower fermentation kinetics due to the presence of mineral-rich molasses components.

4.4 Mineral Content-

Mineral analysis revealed that sodium and potassium concentrations were highest in the control beverage (36.59 ppm Na⁺ and 420 ppm K⁺) and substantially lower in sugar-based beverages. The decline may be attributed to yeast utilization or precipitation during fermentation. Among the sugar sources, palm jaggery beverages showed relatively higher mineral retention compared to refined sugar types, emphasizing the nutritional advantage of unrefined sugar sources.

4.5 Ethanol Production and Purity-

Ethanol concentration varied significantly among the formulations, with the palm jaggery beverage achieving the highest ethanol content (5.94 % v/v) and purity (84 %), followed by sugarcane jaggery and rock sugar variants. The control beverage recorded minimal ethanol (2.91 % v/v) due to limited fermentable sugars. These results confirm that the type and complexity of the sugar source directly affect the fermentation efficiency and ethanol yield, with jaggery-based substrates offering a richer medium for yeast metabolism.

4.6 Phytochemical Composition-

Qualitative phytochemical tests confirmed the presence of flavonoids in beverages prepared with table sugar, rock sugar, and sugarcane jaggery, whereas triterpenoids were absent in all formulations.

The detection of flavonoids indicates partial retention or bioconversion of bioactive compounds during fermentation, contributing to antioxidant and sensory attributes.

4.7 Sensory Evaluation-

The sensory panel revealed that all sugar-based fermented beverages were well accepted (hedonic score = 5), exhibiting pleasant caramel-brown coloration, fruity odour, sour taste, and balanced astringency. The palm jaggery beverage stood out for its distinct vanilla-like aroma and fuller body, likely due to Maillard reaction products and complex volatiles formed during fermentation. Conversely, the control beverage received the lowest score (1), being pungent and bitter—indicating that sugar supplementation is essential for improving flavour and overall palatability.

4.8 Overall Interpretation-

Among the four sugar sources, palm jaggery proved to be the most suitable for producing fermented bottle gourd beverages with superior antioxidant activity, phenolic content, ethanol yield, and sensory quality. The integration of traditional jaggery with bottle gourd fermentation thus provides a functional, nutritionally enriched, and consumer-acceptable beverage. This work also demonstrates the feasibility of utilizing low-cost indigenous materials to create value-added functional drinks with potential nutraceutical applications.

V. CONCLUSION:

The present study successfully developed fermented bottle gourd beverages using different sugar sources and systematically evaluated their biochemical, functional, and sensory characteristics. The results clearly indicated that the type of sugar used in fermentation plays a crucial role in determining the nutritional quality, antioxidant potential, ethanol yield, and consumer acceptability of the final beverage.

Among all the formulations, the palm jaggery—based beverage exhibited the highest antioxidant activity, maximum phenolic and reducing sugar content, moderate ethanol yield, and superior sensory acceptability, making it the most promising formulation. The sugarcane jaggery beverage also showed favourable functional and sensory qualities, highlighting the nutritional advantage of using unrefined sugar sources over refined sugars such as table sugar and rock sugar.

Fermentation significantly improved the functional profile of bottle gourd juice by enhancing antioxidant activity and developing desirable flavour attributes while maintaining product stability. The incorporation of naturally rich sugars like jaggery not only enhanced the nutritional value but also provided a sustainable approach to formulating value-added, health-oriented fermented beverages from locally available agricultural produce.

Overall, this research demonstrates that fermented bottle gourd beverages, especially those prepared using palm jaggery, can serve as functional nutraceutical drinks with potential applications in the food and health industry. Future work may focus on microbial optimization, shelf-life extension, and clinical evaluation to further establish their functional and therapeutic benefits.

VI. FUTURE SCOPE:

The present investigation highlights the potential of fermented bottle gourd beverages as a functional food product; however, several aspects can be explored further to enhance product quality, scalability, and scientific understanding.

6.1 Microbial Optimization-

Future studies can focus on optimizing the fermentation process by exploring different yeast strains or probiotic cultures such as Lactobacillus or Bifidobacterium species to improve fermentation efficiency, nutritional profile, and probiotic potential.

6.2 Shelf-life and Storage Studies-

Long-term stability studies under different temperature and packaging conditions should be conducted to assess the product's microbial safety, sensory stability, and physicochemical changes over time.

6.3 Nutraceutical and Functional Validation-

In-vitro and in-vivo studies may be undertaken to evaluate the antioxidant, antihyperlipidemic, and hepatoprotective effects of the fermented beverages to scientifically validate their health benefits.

6.4 Sensory and Consumer Acceptability-

Wider sensory panels involving different demographic groups can be conducted to understand consumer preferences and tailor the formulation for commercial-scale production.

6.5 Product Diversification-

The bottle gourd-based fermented beverage can be diversified into non-alcoholic or low-alcohol functional drinks, carbonated beverages, or blended formulations with other fruits or medicinal plants to expand its market appeal.

6.6 Industrial and Commercial Application-

Pilot-scale production trials should be undertaken to determine process feasibility, cost-effectiveness, and standardization parameters required for industrial manufacturing and commercialization.

6.7 Nutritional Enhancement-

Further research can explore the fortification of the beverage with vitamins, minerals, or natural extracts to improve its therapeutic value and cater to the growing functional beverage sector.

VII. ACKNOWLEDGEMENT:

We would like to express our sincere gratitude to Guru Nanak Khalsa College of Arts, Science and Commerce (Autonomous) for providing the necessary resources and support during the research project and preparation of this research article.

REFERENCES:

- 1. Barot, A.; Pinto, S.; Balakrishnan, S.; Prajapati, J. P. Composition, Functional Properties and Application of Bottle Gourd in Food Products. Res. Rev.: J. Dairy Sci. Technol., 2015; 4(1): 15-27.
- 2. Saeed, M.; Khan, M. S.; Amir, K.; Bi, J.B.; Asif, M.; Madni, A.; Kamboh, A. A.; Manzoor, Z.; Younas, U.; Chao, S. *Lagenaria siceraria* fruit: A review of its phytochemistry, pharmacology, and promising traditional uses. Front Nutr., 2022; 9: 927361.
- 3. Ahmad, M.; Ahmad, I.; El-Chaghbay, G.; Rashad, S. Nutritional and Medicinal Potential of Bottle Gourd (Lageneria siceraria): A Mini Review. Egyptian J. Bot., 2022; 62(1): 1-10.
- 4. Minocha, S.; Tiwari, A.; Gandhi, S.; Sharma, A.; Gupta, A. AN OVERVIEW ON *Lagenaria* siceraria (BOTTLE GOURD). J. Biomed. Pharm. Res., 2015; 4(3): 4-10.
- 5. Fruitique. Loki (Bottle Gourd) [Photograph]. Fruitique; c2021 [cited 2024 July 27]. Available from: https://fruitique.in/products/bottle-gourd-loki

- 6. AgriBegri. Bottle Gourd Gardening For Beginners, How To Start [Internet]. AgriBegri; [cited 2024 July 27]. Available from: https://blog.agribegri.com/en/blog/bottle-gourd-gardening-for-beginners-how-to-start
- 7. DunleathGarden. Female bottle gourd flower close-up [Photograph]. Dunleath Community Garden; 2020 Jul 8 [cited 2024 July 27]. Available from: https://dunleathgarden.wordpress.com/2020/07/08/brinta-helps-female-bottle-gourd-flowers-get-the-pollen-they-need-to-produce-fruit/
- 8. Naati Grains. Bottle gourd seeds [Photograph]. Naati Grains; [cited 2024 July 27]. Available from: https://www.naatigrains.com/buy-bottle-gourd-seeds-online
- 9. Attar, U. A.; Ghane, S. G. In vitro antioxidant, antidiabetic, antiacetylcholine esterase, anticancer activities and RP-HPLC analysis of phenolics from the wild bottle gourd (*Lagenaria siceraria* (Molina) Standl.). South Afr. J. Bot., 2019; 125: 360-370.
- 10. Devi, S. R.; Kumari, T.; Deka, S. C. Extraction of dietary fiber and phytochemicals from bottle gourd seeds (*Lagenaria siceraria*), its physicochemical properties and application in food model. Food Chem. Adv., 2023; 2: 100252.
- 11. Naysadorai, K. A.; Bellary, S. Bottle gourd (*Lagenaria siceraria*) toxicity: A rare case report. Clin. Med., 2024; 24 (suppl.): 100077.
- 12. Xiao, F.; Xu, T.; Lu, B.; Liu, R. Guidelines for antioxidant assays for food components. Food Frontiers, 2020; 1: 60-69.
- 13. Shimamura, T.; Sumikura, Y.; Yamazaki, T.; Tada, A.; Kashiwagi, T.; Ishikawa, H.; Matsui, T.; Sugimoto, N.; Akiyama, H.; Ukeda, H. Applicability of the DPPH Assay for Evaluating the Antioxidant Capacity of Food Additives Inter-Laboratory Evaluation Study-. Anal. Sci., 2014; 30(7): 717-721.
- 14. Dr. Mahesha, H.B. Estimation of Protein by Lowry's method. Yuvaraja's College, [date unknown]. [cited 2024 July 27] Available from: https://hbmahesh.weebly.com/uploads/3/4/2/2/3422804/estimation_of_protein_by_lowrys_method.pdf
- 15. Singh, A. Estimation of protein, oil, and sugar content in maize (Zea mays) seeds. SRM University, [date unknown]. [cited 2024 July 27] Available from: https://www.researchgate.net/profile/Arvind_Singh56/post/Which_is_the_economical_and_easy_method_to_estimate_protein_oil_and_sugar_content_in_maize_corn_seeds/attachment/59d63c127_9197b8077999105/AS%3A413819290570753%401475673615545/download/1.pdf
- 16. Mula, M. Determination of reducing sugar by DNS method. [date unknown]. cited 2024 July 27] Available from: https://www.scribd.com/document/539385004/Determination-of-reducing-sugar-by-DNS-method
- 17. Author unknown. Estimation of reducing sugars by dinitrosalicylic acid method. [date unknown]. [cited 2024 July 27] Available from: https://fac.ksu.edu.sa/sites/default/files/445_lab_2.pdf
- 18. vlab.amrita.edu,. (2012). Flame Photometry. Retrieved 20 June 2024, from https://vlab.amrita.edu./?sub=2&brch=294&sim=1351&nt=1
- 19. Author unknown. How to make bitter gourd wine. Delishably, [date unknown]. [cited 2024 July 27] Available from: https://delishably.com/beverages/How-to-make-BITTER-GOURD-WINE
- 20. Savanur, P. V. Phytochemical study of Usheera (*Vetiveria zizanioides* (Linn) Nash). Int. J. Ayurveda Herb. Med., 2017; 7(3): 2605-2628.
- 21. Rao, A.; Kumari, S.; Laura, J. S.; Dhania, G. Qualitative Phytochemical Screening of Medicinal Plants using Different Solvent Extracts. Oriental J. Chem., 2023; 39(3): 621-626.
- 22. Solution-Pharmacy. Chemical Identification Test for Steroids & Triterpenoids: GPAT, Pharmacist & other Exam Exclusive [Internet]. YouTube, 2021. [cited 2024 July 27] Available from: https://youtu.be/ZWL7esNFbmc
- 23. Virginia Polytechnic Institute and State University; Virginia State University; U.S. Department of Agriculture. *What do I need to know to sell Fermented Vegetables at the farmers market?* Publication FST-308P (FST-365P) [Internet]. 2020 [cited 2024 July 22]. Available from: https://share.google/uRUl7jhObz1i5nZ3A

- 24. Enartis S.A. Wine stability: South Africa newsletter [Internet]. [place unknown]: Enartis; Oct 2021 [cited 2024 Jul 13]. Available from: https://www.enartis.com/wp-content/uploads/2021/10/Enartis_Newsletter_Wine_Stability_SA.pdf
- 25. Iowa State University Extension and Outreach. FS 40: Wine Faults Series [Internet]. Ames (IA): Iowa State University; [date unknown; cited 2024 Jul 13]. Available from: https://www.extension.iastate.edu/wine/wp-content/uploads/2021/09/FS40.pdf
- 26. Australian Wine Research Institute. Measurement of cold stability of wine [Internet]. The Australian Wine Research Institute; [cited 2024 Jul 13]. Available from: https://www.awri.com.au/industry_support/winemaking_resources/laboratory_methods/chemical/cold_stab/
- 27. Miranda J. Total phenolics by Folin Ciocalteu [Internet]. YouTube; 2023 [cited 2024 July 27]. Available from: https://youtu.be/5f1zWp2UwyM
- 28. Bevanda, A. M.; Ivankovic, A.; Talic, S.; Stipanovic, A.; Petrovic, D. Determination of ethanol in wine and distilled alcoholic beverages using a smartphone as a detector. Int. J. Comput Theor Chem. 2021; 9(2): 43-7.
- 29. Pagliarini, E.; Laureati, M.; Gaeta, D. Sensory descriprors, hedonic perception, and consumer's attitudes to Sangiovese red wine deriving from organically and conventionally grown grapes, Front. Psychol., 2013; 4: 896.
- 30. Joshi, V.K.; Kumar, N. Optimization of low alcoholic bitter gourd apple beverage by applying Response surface methodology (RSM). Intl. J. Food Ferment. Technol., 2015; 5(2): 191-199.

