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Physico-biochemical study of sugarcane jaggery under different cultivation and preparation methods

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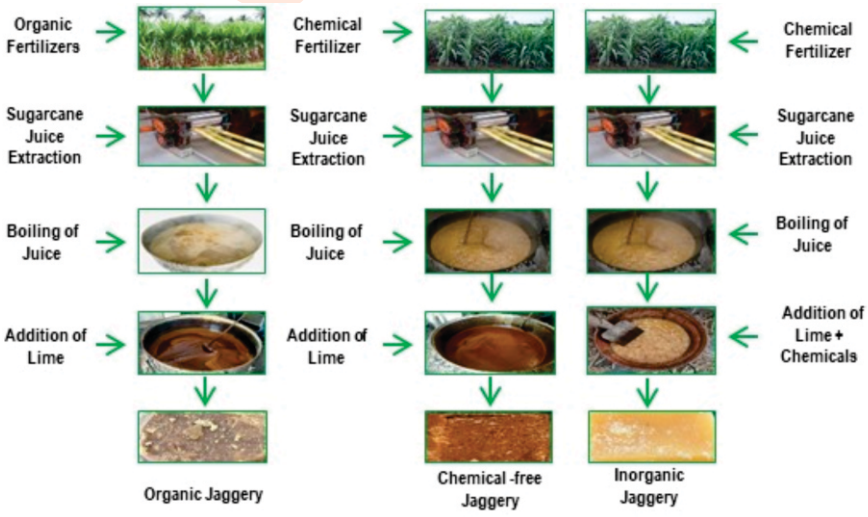
Abstract

Aim: The present study was undertaken to compare the physico-biochemical and microbial qualities of organic, chemical-free and inorganic jaggery and the changes in microbial count during storage.

Methodology: Jaggery was prepared by evaporating of sugarcane juice with or without addition of chemicals from organically or inorganically cultivated sugarcane. These organic, chemical-free and inorganic jaggery were analysed for physico-chemical, bioactive compounds and microbial properties.

Results: The present study results that organic and chemical-free jaggery comes under Grade I, while inorganic jaggery comes under Grade II. The organic jaggery and chemical-free jaggery recorded 61% and 53%more total polyphenol content; 51% and 46% more antioxidant potential and 6% and 10% more total flavonoids compared to inorganic jaggery respectively. However, the shelf-life of organic jaggery was less compared to inorganic jaggery due to high moisture content.



Interpretation: The present study revealed significant difference in quality parameters in all three jaggery samples. Though, the inorganic jaggery was brighter in color with low microbial load, it falls short of beneficial components that are lost due to addition of chemicals during processing. Hence, organic jaggery followed by chemical-free jaggery can be recommended to both jaggery farmers and consumers for premium price and nutritional health benefit.

Key words: Jaggery, Quality analysis, Sugarcane

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Introduction

Jaggery is an eco-friendly traditional Indian sweetener. Jaggery is produced worldwide annually across 25 nations.Out of the entire jaggery production in the world, 70% production (approximately 10 million tons) takes place in India alone (Venkatesan *et al.,* 2024). India is world's leading jaggery producer and user. The other jaggery producing countries are Brazil, Thailand, Australia, Germany, Mexico, *etc.,* and the main importing countries are USA, China, Indonesia, Malaysia, Japan. Hence, jaggery is recognized as one of the chief traditional agricultural export products in India (Revathy *et al*., 2021). In India, majority of jaggery is being produced during November to April (Uppal and Sharma, 1999). In the year 2019-2020, India exported approximately 341155.34 MT of jaggery and its products worth USD 227.90 million (Singh *et al.,* 2021). The chemical composition of jaggery comprises of sucrose (65-85%), reducing sugars including glucose and fructose (10-15%), non-sugars such as calcium (0.4%), magnesium and phosphorous (0.045%), protein (0.25%) and fat (0.05%) (Rao *et al.,* 2007).

The nutritional qualities of jaggery helps in purification of blood, prevent rheumatic and bile disorders, helps in curing jaundice (Singh *et al.,* 2013). Jaggery has numerous therapeutic properties like antioxidant activity, antimicrobial activity, cytoprotective, and neuroprotective activities (Barrera *et al.,* 2020; Nayaka *et al.,* 2009; Sharma and Sharma, 2012) due to the presence of different bioactive compounds. Though jaggery possess various nutritional and medical qualities, it is not available in the market in pure form. Majority of the jaggery existing in the market ispale yellow to dark brown in color with varying quality parameters. The major attributes that govern the jaggery quality are sucrose, reducing sugars, moisture content, and presence of chemicals. The marketability of jaggery mainly depends on color, texture and shelf-life of jaggery. The consumers usually select yellow color jaggery for consumption. It is reported that during jaggery preparation, addition of chemicals imparts pale yellow color to jaggery which has detrimental effect on health. In the view of increasing health consciousness of the consumers, the focus of interest has shifted towards organic and chemical-free products.

The jaggery available in the market is sold under different names such as organic jaggery, chemical-free jaggery. Based on cultivation and preparation methods, the jaggery can be categorised into three types: Organic jaggery, Chemical-free jaggery and Inorganic jaggery. In organic jaggery, sugarcane is grown organically without making use of chemical fertilizers and jaggery is prepared from this sugarcane with no chemicals. The chemical-free jaggery is prepared from sugarcane crop cultivated with recommended dose of fertilizers (Lakshmi *et al*., 2011) and jaggery is prepared from this sugarcane without using any chemicals. The recommended dose of N-P_2O_5-K_2O for sugarcane plant crop is 112-100-120 kg ha^{-1} (Lakshmi *et al*., 2011). Nitrogen is applied in two equal split doses at 45 and 90 days after planting, whereas entire Phosphorus and 50% Potassium is applied at the time of planting and the remaining 50% potassium is applied at 90 days after planting.

At present, the scientific study about the effect of cultivation and jaggery preparation method on quality of jaggery is no where documented. Hence, the present study was undertaken to compare the bio-chemical, bioactive compounds and microbial qualities of these three types of jaggery. Three jaggery samples under study were sugarcane cultivated organically and jaggery made without adding chemicals, sugarcane grown using chemical fertilizers and jaggery prepared without using any chemicals and sugarcane cultivated using chemical fertilizers and jaggery produced by adding chemicals.

Materials and Methods

Organic jaggery preparation: Production of organic jaggery depends on agronomic management practices followed in sugarcane cultivation and jaggery preparation process. Organic jaggery is natural, pure, and chemical-free and free from all impurity. The Sugarcane Var.87A298 was cultivated without application of expensive inorganic fertilizers, plant protection chemicals. The agronomic management practices include replacement of chemical fertilizers by Dhravajeevamrutham and Ghanajeevamrutham which were prepared with the locally available resources. Jeevamrutham is an organic fertilizer and a great replacement of chemical fertilizers. It is a good source of biomass, natural carbon, nitrogen, phosphorous, calcium and other nutrients, essential for plant growth and development. The microorganisms present in the soil helps to increase the soil fertility and crop productivity (Kaur, 2020). Ghanajeevamrutham is prepared using desi cow dung, besan flour, jaggery whereas Dhravajeevamrutham is prepared from cow dung, cow urine, jaggery, pulse flour and clean water. After attaining maturity, the cane is harvested and jaggery is prepared without using any chemicals. The various unit operations for jaggery preparation include pre-cleaning of canes, juice extraction, and clarification, boiling of juice, concentration, cooling and moulding. Among all the operations, clarification of the juice is essential to maintain the quality standards. The fresh sugarcane juice contains colloidal inorganic and organic suspensions, non-sugars which are to be removed during jaggery making by clarification process to impart golden yellow colour to jaggery (Gangwar *et al.,* 2015). During jaggery preparation no chemicals are added. However, Calcium hydroxide (milk of lime), which was prepared from natural source was added as basic clarificant to remove impurities as scum (Fig.1). Calcium hydroxide was procured from the local market which was prepared from sea shells as they are natural source of calcium carbonate.

Chemical-free jaggery preparation: Sugarcane Var.93A 145 was cultivated at Regional Agricultural Research Station (RARS), Anakapalle as per recommended dose of chemical fertilizers (ANGRAU). This cane was utilized for making jaggery without using chemicals. Mature sugarcanes were crushed to obtain juice filtered using steel mesh to remove coarse impurities and used for

preparing jaggery. Milk of lime was added as a basic clarificant to separate the scum and regulate the pH of the juice. Initial pH of the sugarcane juice was 5.2 and lime was added during boiling until the pH reached to 5.8. (Rao *et al.,* 2007). The juice was boiled until the water in the sugarcane juice gets evaporated to get concentrated product of brix 85° and temperature of 118 °C was attained. The concentrated jaggery syrup was unloaded, cooled, and moulded into desired shapes. The jaggery thus obtained was utilized for quality analysis.

Inorganic Jaggery: In this process, jaggery is prepared from sugarcane which is cultivated using chemical fertilizers. During jaggery preparation, farmers add soda, lime and sodium hydrogen sulphite called as hydros to sugarcane juice for improving the colour. The organic and inorganic jaggery samples were collected from jaggery farmers where sugarcane is cultivated and jaggery is prepared by them whereas chemical-free jaggery was prepared at Regional Agricultural Research Station, Anakapalle from the sugarcane cultivated at the farm. These samples were used for the analyses of physico-chemical, bioactive compounds and microbial counts.

Estimation of physico-chemical properties

The Colour transmittance of jaggery was determined according the method described by Mandal *et al.* (2006). The moisture content of jaggery samples were determined by gravimetric method as per AOAC (2000). About 5 gof jaggery sample was taken in a aluminum box and the sample is dried in oven at 70 °C for 6-8 hr. The sample is taken out and cooled in desiccator and loss in weight is reported. The process was repeated until a constant weight was obtained. The loss in weight was calculated as moisture content of the sample expressed in percentage dry basis. The percent sucrose of samples was recorded using polarimeter (Antanpaar, Model: MCP 500 Sucromat). The percent reducing sugars are estimated by Lane and Eynon method (1923) using Fehling A and Fehling B solutions according to the principle of reduction of Fehling's solution by reducing sugars (Lane and Eynon, 1923).

Total polyphenol content of jaggery samples were estimated using Folin-Ciocalteu method (Singleton *et al.,* 1999). About 100 µl of 5% of jaggery sample was added to 900µl of distilled water and 1 ml of FCR reagent (1:2 v/v), thereafter 10% sodium carbonate was added and the solution was mixed with a vortex meter and kept in dark for 30 min for incubation. The absorbance of the aliquot was read at 765 nm on a Spectrophotometer (Make: Elico, Model: SL27VIS spectrophotometer). The standard curve was prepared using gallic acid and line of equation was made. The total polyphenol content was expressed in gallic acid equivalent in µg ml^{-1} jaggery.

Total flavonoid content of jaggery samples were estimated as described by Chang *et al.* (2002) using aluminium chloride. About 200 µl of 5% jaggery sample was made up to 2 ml with distilled water. To it 100 µl of 10% sodium chloride and 100 µl of 0.5 ml of 1M potassium acetate were added followed by vortex. The solution is then incubated for 30 minutes and using spectrophotometer, the absorbance was recorded at 415 nm. The standard curve using quercetin at different concentrations was prepared, and the total flavonoid content of the sample was expressed as quercetin equivalent (QE) µg ml^{-1} jaggery.

The DPPH radical scavenging activity of jaggery was quantified following the method of Yamaguchi *et al.* (1998). Accordingly, a sample aliquot of jaggery at l0-50 µg ml^{-1} and standard antioxidant at 2-10 µg ml^{-1} was made up to 200 µl using distilled water. 1 ml of 0.1 mM DPPH was added. The solution was kept in dark at room temperature for 20 min and the absorbance was noted against control at 517 nm. The scavenging activity was calculated and the result was expressed as an effective concentration (EC_{50}) for 50% DPPH radical scavenging activity.

DPPH Radical Scavenging Activity (%)=$[(A_{control}-A_{sample})/A_{Control}] \times 100$

The hardness of jaggery was determined by Textural analyser (Model: TAXT PLUS) Made: Stable Micro Systems). For determining of hardness of jaggery, a cylindrical probe P/2 was used and the machine was calibrated using 50 kg load cell. The jaggery sample was kept on a raised platform to imitate the experiment. The plot between the force and time was made by software. The peak of the plot represents the fracture and the sequence of peaks appeared as compression progressed. The highest peak value showes the hardness of the sample (Kumar *et al.,* 2013). The microbial analysis of jaggery samples was evaluated for aerobic mesophiles (AM), fungal counts using the serial dilution pour plate method (APHA, 2015). Potato Dextrose Agar and Nutrient Agar were used as the growth media for the fungaland AM respectively. All the media were procured from HIMEDIA®, India (HiMedia Laboratories Pvt. Ltd., India).

The corresponding incubation conditions for the microbial growth (AM and fungal) were 37 °C/24–36 hr, 37 °C/18–24 hr respectively. After incubation, microbial colonies were counted and noted as colony forming units (CFU). The minimum detection level for sugarcane juice was 1 CFU ml^{-1} (Sreedevi and Kuruba, 2019). The experiments were performed in triplicate and the mean values were expressed as log 10 CFU ml^{-1} of the sample. During shelf-life studies, microbial analysis was done for 3 months at 30 days interval for three jaggery samples.

Statistical Analysis: The jaggery samples from three different categories was analysed for quality parameters and analysis was done in three replications. The average of three reading was calculated and expressed as mean ± standarddeviations. SPSS software version 2.0 (IBM SPSSStatistics, USA) was used to carry out the significance testsviz. Fisher's least significance difference (LSD) andTukey's HSD test at 95% confidence interval for all the parameters under study.

Results and Discussion

The quality analysis of different types of jaggery under study *viz.* organic, chemical-free jaggery and inorganic jaggery were analyzed. The sucrose and percent reducing sugars are the major factors that govern the quality of jaggery. A significant difference was observed in sucrose and reducing sugars percent among all the three jaggery samples. For Grade I jaggery, the jaggery should contain high sucrose percent (more than 80%) and low reducing sugars (less than 10%)as per the criteria requirements given in Table 1. The sucrose percent was more than 80% and the reducing sugar percentage was less than 10% in organic and chemical-free jaggery (Table 2) indicating that these samples come under Grade I according to Prevention of Food Adulteration (PFA) Act (1954). In inorganic jaggery the sucrose content was 78.6% and the reducing sugar was 11.4%, thus all under Grade II jaggery.Higher value of reducing sugars in jaggery sample indicates hygroscopic nature and is prone for microbial infestation. It was reported that low reducing sugar is preferable for goodquality jaggery as it is generally less hygroscopic (Singh *et al.,* 2019). The moisture content of organic jaggery was high (6.92%), followed by inorganic jaggery (5.97%) and chemical-free jaggery (5.88%).

Higher the moisture content of the product, the higher will be the chances for microbial infestation and lower will be the shelf-life.The marketability of jaggery is largely dependent on the color, as grading of jaggery in India is carried out by its physical color. A significant colour difference was observed in organic and inorganic jaggery. The color of jaggery in terms of percent transmittance was high in inorganic jaggery representing light yellow compared to organic and chemical-free jaggery (Fig. 1), which might be due to the addition of chemical clarificants such as sodium hydrogen sulphite commonly known as hydros during jaggery making. It was reported that hydros added as a chemical clarificant is a powerful reducing agent and reduces colored quinones into colour less phenols (McEvily *et al.,* 1992; Lambrecht, 1995). The jaggery prepared using chemicals exhibited a pale yellow color which temporarily enhances the physical appearance but indiscriminate use of this chemical resulted in deposition of harmful chemicals. The hydros used in food and beverage industries helps in inhibiting browning reactions, such as phenol oxidation and Maillard reactions (Verma *et al.,* 2019).

The bioactive compounds of jaggery under study were evaluated for total phenolic content, total flavonoids and antioxidant activity. Polyphenols present in non-centrifugal sugars have proved to enhance the health positively (Jaffe, 2012). A significant difference was observed in polyphenolic content among the three samples. The phenolic content of organic jaggery (4080 µg GAE g^{-1}) and chemical-free jaggery (3862 µg GAE g^{-1}) were 61% and 53% more than inorganic jaggery (2522 µg GAE g^{-1}) respectively. Similar result of phenol content was recorded in jaggery (3837 µg GAE g^{-1}) followed by brown sugar (372 µg GAE g^{-1}), white sugar (31.5 µg GAE g^{-1}), and refined sugar (26.5 µg GAE g^{-1}) by Sharifi-Rad *et al.* (2023). The addition of chemicals during jaggery making resulted in loss of the desired components such as polyphenols, minerals and vitamins as reported by Hussain *et al.* (2012).

The total flavonoids in the jaggery samples under study varied from 479 to 531µg QE g^{-1}. Higher flavonoid content (531 µg QE g^{-1}) was recorded for chemical-free jaggery whereas lowest value of (479 µg QE g^{-1}) was recorded for inorganic jaggery. The addition of hydros during jaggery making in inorganic jaggery has reduced the polyphenols and flavonoids. Similar results of reduction in polyphenols and flavonoids were reported for jaggery prepared using chemicals as clarificant (Sreedevi *et al.,* 2022). The radical scavenging potential of jaggery samples were expressed as EC_{50} values (half-maximal effective concentration). Lower the EC_{50} values of an antioxidant, more will be the free radical scavenging potentiality. Out of the three samples under study, organic jaggery recorded lowest EC_{50} value (6.49 mg ml^{-1}), followed by chemical-free jaggery (7.10 mg ml^{-1}) and inorganic jaggery (13.3 mg ml^{-1}). Similar result of EC_{50} of 7.81 and 59.38 µg ml^{-1} was reported by Nayaka *et al.* (2009) for jaggery and brown sugar, respectively, in the DPPH scavenging assay. Free radicals occur because of normal processes of energy production inside the body of human being or may be exogenous (Mititelu *et al.,* 2020). To neutralize them, natural antioxidants are a good alternative (Islam *et al.,* 2021a; Islam *et al.,* 2021b). Hence, organic jaggery with low EC_{50} act as strong antioxidant potential in scavenging assay.

The hardness of jaggery affects the quality of jaggery and its shelf-life. The lower hardness values in jaggery represent softness with low shelf-life. A significant difference in hardness

Table 1: Characteristic requirements for grading of Indian jaggery

Characteristic requirements		
	Grade I	**Grade II**
Sucrose (on dry basis), percent by mass, min	80	70
Reducing sugars (on dry basis), percent by mass max	10	20
Moisture percent by mass; max	5	7
Sulphated ash (on dryy basis), percent by mass; max	3.5	5

Source: Indian standard cane gur (jaggery)—specification IS 12923:1990

Table 2: Quality analysis of organic, chemical-free and inorganic jaggery

Quality Parameter	Organic jaggery	Chemical-free jaggery	Inorganic Jaggery
Physico-chemical analysis			
Sucrose (%)	86.3±2.1[b]	84.2±2.5[b]	78.6±2.5[a]
Reducing sugar (%)	3.6±0.8[a]	5.8±0.6[b]	11.4±0.7[c]
Moisture (%)	6.9±0.3[b]	5.8±0.2[a]	5.9±0.2[a]
Color (Transmittance %)	52.1±5.1[a]	51.8±4.7[a]	58.3±4.5[b]
Hardness (N)	137.3±11[a]	215.7±17[c]	196.1±12[b]
Bioactive compounds			
Total phenolic content (μg GAE g^{-1})	4080±25[c]	3862±31[b]	2522±18[a]
Total flavonoids (μg QE g^{-1})	510±12[b]	531±14[b]	479±11[a]
Ec_{50} (mg ml^{-1})	6.4± 0.25[a]	7.1±0.31[b]	13.3±0.5[c]

Alphabets in small letters (a, b and c) in the superscripts denote that the mean values are statistically different within the row at $p< 0.05$. Values are reported as mean of three replicate ± S.D.; ND- Not detected

Organic Jaggery | Chemical free Jaggery | Inorganic Jaggery

Fig. 1: Jaggery prepared under different cultivation and preparation methods.

values was noted among the three samples. Highest hardness value (215 N) was recorded for chemical-free jaggery, followed by inorganic jaggery (196 N) and organic jaggery (137 N). Higher moisture content in organic jaggery supports the lower hardness value compared to other jaggery samples. Similar hardness value of jaggery samples ranging between 290 N to 400 N was reported by (Sahu *et al.,* 2024).

The microbial analysis of jaggery samples were carried out for total bacterial and fungal count (Table 3). Initially, no bacterial and fungal count was observed in the three jaggery samples. During 3 months storage, it was observed that bacterial count had increased in all the samples with highest count recorded for organic jaggery and lowest for inorganic jaggery. It was already reported that the microbial counts were more in normal jaggery compared to hydros-treated jaggery samples (Verma *et al.,* 2019). The addition of hydros during jaggery making will help in the preservation of jaggery (Ravindra *et al.,* 2016). Sulphites formed during hydros treatment are proved as antimicrobial agents in various foods (Lambrecht, 1995). Hence, less bacterial count was recorded for inorganic jaggery. No fungal count was reported for chemical-free and inorganic jaggery up to 30 days. However, after 30 days of storage, the fungal count was observed to be more in organic jaggery compared to inorganic and chemical-free jaggery. Hence, organic jaggery with high moisture content and low hardness value had low shelf-life compared to chemical-free and inorganic jaggery. Overall, the organic jaggery and chemical-free jaggery recorded 61% and

Table 3: Shelf-life studies of organic, chemical-free and inorganic jaggery

Jaggery	Total bacterial count (cfu ml^{-1})				Fungal count (cfu ml^{-1})			
	0 Day	30 Day	60 Day	90 Day	0 Day	30 Day	60 Day	90 Day
Organic jaggery	ND	2.92±0.15^{a}	4.32±0.25^{b}	5.15±0.31^{c}	ND	1.00±0.10^{a}	2.00±0.10^{b}	2.30±0.15^{c}
Chemical-free jaggery	ND	2.65±0.15^{a}	4.25±0.25^{b}	4.78±0.28^{b}	ND	ND	1.3±0.10^{a}	2.00±0.15^{a}
Inorganic Jaggery	ND	2.80±0.16^{a}	3.96±0.24^{b}	4.60±0.29^{c}	ND	ND	ND	1.00±0.10^{a}

Alphabets in small letters (a, b and c) in the superscripts denote that the mean values are statistically different within the row at $p< 0.05$, Values are reported as mean ± standard deviation (N=3); ND- Not detected

53% more total polyphenol content; 51% and 46% more antioxidant potential and 6% and 10% more total flavonoids compared to inorganic jaggery respectively. However, keeping in view of lower shelf-life, it was recommended to market the organic jaggery within short time. Hence, it was suggested to produce organic jaggery that need to be marketed within 3 months as it was prone to microbial infestation. After covid pandemic,more emphasis and interest is shown towards usage of chemical-free, nutritional rich products by the populace worldwide. This paves a way for promoting of organic or chemical-free jaggery. Now-a-days, there is great demand for organic foods.Particularly in developed countries, majority of consumers prefer organic food to be safer and healthier than inorganically cultivated food (Funk and Kennedy, 2016). In this context, the deep study on quality aspects of organic, inorganic and chemical-free jaggery will help the consumers to choose a better quality jaggery.

The research findings can be helpful for future research on the study of residual effects of chemicals on jaggery. The organic or chemical-free jaggery has a much better nutritional value than inorganic jaggery and fetches a good price to jaggery farmers. However, further studies need to be done to access the cost economics of these three types of jaggery. The comparative analysis of organic, chemical-free, and inorganic jaggery reveals that organic jaggery followed by chemical-free jaggery have superior quality compared to inorganic jaggery in terms of bioactive compounds like polyphenols and flavonoids, which contribute to better antioxidant activity. Though inorganic jaggery presented light-yellow appearance, enhanced by chemical additives like sodium hydrogen sulphite, it falls short of nutrition qualities due to loss of beneficial components during processing. Given the increasing demand for natural, chemical-free products, organic and chemical-free jaggery are recommended as better alternatives, though further studies are needed to ensure market viability and cost-economics.

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