DEVELOPMENT OF FLAVOUR ENRICHED JAGGERY CUBES

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Abstract: Sugar industry is the second largest agro-base industry and contributes significantly to the socio-economic development of the nation. In the present investigation blend of ginger lemongrass oleoresin was taken to provide health benefits with taste to satisfy the consumer need and demand. In the present investigation the different concentration (0.6, 0.8 and 1.0%) of blend of ginger lemongrass oleoresin was used to optimize flavor enriched jaggery cubes. Among all 0.8 % concentration of flavor was selected and evaluated for its antimicrobial studies. The result of antimicrobial studies revealed that the selected concentration of oleoresin enriched sugarcubes are most effective for gram positive bacteria like *Bacillus Substilus* followed by *E.coli* and gram negative bacteria. Storage studies of developed cubes were carried out at different conditions like, storage temperature (20-25˚C, 40-45˚C) and packaging material (HDPE,LDPE). Further bioactive compounds were checked and observed slight degradation in phenolic compounds and antioxidant activity during storage with respect to time. The present study indicated that the development of flavor jaggery cubes will be boon for sugar producer and processing industries.

IndexTerms **- Jaggery Cubes, Antioxidant Activity, Total Phenolic Content, Ginger lemongrass flavour**

I. INTRODUCTION

Sugar industry is plays a leading role in global market after Brazil, produces nearly 15 and 25 per cent of sugar and sugarcane respectively. Worldwide India produces more than 70% sugarcane, out of which 53% is processed into white sugar, 36% processed into jaggery and khandsari, 3% for chewing as cane juice, and 8% as seed cane to full fill the need as a sweeteners. Sugar industries are providing livelihood to more than 50 million farmers and their families. Therefore sugar cane industries are economic backbone of the nation (Kshirsagar, 2012).

As mention above, sugar cane is processed into many products, among them jaggery is a natural sweetener made by the concentrating a sugarcane juice to a certain level. It is a rich source of iron and therefore consumption increases the level of hemoglobin in the blood. Along with iron, jaggery is a prominent source of minerals and vitamins. Therefore jaggery is the healthiest sugar in the world (Kshirsagar, 2012). Due to its high nutritional value jaggery industries occupy a prominent place in the sugar economy and it consumes 20.36% of the total sugarcane grown in India. To develop light colored jaggery, herbal clarificant (deola extract @ 45 g/100 kg juice) is use to eliminate impurities from colloidal suspension and coloring compound from accumulation process. The structure of jaggery is more complex than sugar, and made up from longer chains of sucrose. Jaggery digest slowly as compare to sugar and it releases energy gradually to keeps body energetic for longer time. Therefore utilization of jaggery in any product will be achievement for processing industries.

Flavoured sugar cubes like orange, blossom, rosewater, lemon and cardamom are available in the market, can be used in the beverages to sweeten the drinks but does not provide health benefits and appealing taste to the consumer. Scanty of literature is available on the utilization of herbs and spices for preparation of flavoured cubes. Similarlly no reports are available on the utilization of jaggery, hence scientists are looking for innovation. Consumers are also in quest of natural foods and natural preservatives for healthier lifestyles and natural ways of preventing ailments. Indian herbs posses multifarious properties and provide tons of health benefits if you consume**.** The hypothesis behind this

work was to prepare flavoured jaggery cubes for beverages like tea and coffee, as they are most favourite beverage and fulfill the thirst quenching properties of the consumer. It can be instant energy source for hot (tea and coffee) and cold (Ice tea) beverages. This product will be boon to the processing industry. Ginger and lemongrass are the important tea ingredients posseses huge benefits to the consumer.

Ginger (*Zingiber officinale* Rosco), member of *Zingiberaceae*, it has been used for over 2000 years (Stoilova *et al.* 2007; Hasan *et al.* 2012). The distinct yellow, pungent, aromatic rhizome is the plant's organ that confers its value to the spice and the source of oleoresin and the essential oil. The chemical studied of ginger found that it has over 400 different constituents. The major pungent compounds from thelipophilic rhizome extract have yielded potentially active gingerols, which can be converted to shogaols, zingerone and paradol. Gingerols known as phenolic ketones can be converted to shogaols, zingerone, and paradol (Rahmani *et al.* 2014) which produce the "hot" sensation in the mouth (Aly *et al.* 2013). The important compounds are carbohydrates (50–70%), lipids (3–8%), phenolic acids, and terpenes in ginger rhizomes **(**Mele, 2019). Gingerol is responsible for its characteristic aroma and taste.

Lemongrass is known as *Cymbopogon citratus* belongs to the family *Poaceae*, contains several important bioactive compounds which are useful in many health diseases. Essential oil is one of the important components of lemon grass extracts and its applications include co-ingredients for perfumes and cosmetics. Lemongrass oil has been found to contain up to 75-85% citral, borneol, estragole, methyl eugenol, geranyl acetate (3,7-dimethyl-2,6-octadiene-1-ol acetate), geraniol (some species higher in this compound than citral) (Anggraeni *et al.* 2017). The different compound in lemongrass possesses various pharmacological activities such as anti-amoebic, anti-bacterial, anti-diarrheal, anti-fungal and anti-inflammatory properties (Hasim *et al.* 2015).

As discussed, ginger and lemongrass are having tons of health benefiting properties. Hence small attempt was taken to provide health benefits with taste to satisfy the consumer need and demand. Tea without ginger and lemongrass can't imagine. Therefore the development of flavor cubes using blend of ginger and lemongrass oleoresin will be the good idea to prepare instant product.

II. MATERIAL AND METHODOLOGY:

2.1. RAW MATERIALS

Organic jaggery was procured as per the requirement of product characteristics from the local market of loni kalbhor. The **b**lend of ginger-lemongrass oleoresin was purchased from International flavor and fragrances (IFF) India pvt.ltd, Chennai.

2.2 Packaging material

Low density polyethylene and high density polyethylene bags were selected to stored flavoured cubes during storage. Packaging material was purchased from local market of Pune.

2.3 Chemicals

Analytical grade chemicals i.e sodium bicarbonate (Na_2CO_3), Sodium hydroxide (NAOH), Hexane, Folin-Ciocalteu reagent (Sigma-Aldrich pvt.ltd, Pune), DPPH (2,2-diphenyl-1-picrylhydrazyl) (Sigma – Aldrich pvt.ltd,) reagent was purchase from marshal Laboratories Private Limited, pune.

2.4 Strains of microorganisms

To check the antimicrobial activity of samples most common gram positive and gram negative strains like *Bacillus cereus (B. cereus), Escherichia coli (E. coli)* and fungal strain *Aspergillus Niger (A.niger),* were obtained from microbiology lab of MIT College of Food Technology, Pune.

2.5 Chemical analysis

2.5.1 Total phenolic content

Total phenolic contents of all sample extracts were determined using Folin-Ciocalteu reagent as described by Singlaton and Rossi (1965) with slight modification. Extracted 100 µl samples were inserted into different test tube and mixed thoroughly into 900 µl Folin-Ciocalteu reagent (Pre-dilute with distilled water, 10 times) and makeup the volume up to 5 ml with water. After 5 mins, 750 µl of NA_2CO_3 of 7.5% sodium carbonate (NA_2CO_3) was added and allowed to react for 90 min at room temperature (20-25˚C). The absorbance was measure at 765 nm using spectrophotometers. Standard curve of gallic acid solution (20, 40, 60, 80, 100 and 120µl) was prepared using the similar procedure. Total phenolic contents were expressed as mg gallic acid equivalents (GAE)/g of crude extract from the gallic acid calibration curve using the equation y=0.0012x-0.007 (R^2=0.995). All samples were analyzed in triplicate.

2.5.2 Antioxidant analysis

Free radical scavenging activity of sample was determined by 2,2-Diphenyl-1-picrylhydrazil radical (DPPH) and expressed % radical scavenging activity. DPPH solution (1 mg/ml) was made by dissolving DPPH in methanol. DPPH solution (100 µl) was diluted to 5 ml and absorbance was taken at 517 in UV-Spectrophotometer. Five different concentrations (0, 20, 40, 60, 80, 100**µl**) of ascorbic acid were taken and plotted standard curve against absorbance. The extract (100µl) was made by dissolving required oleoresin in methanol then it was added with 100µl of 1mg/ml of DPPH solution and was incubated at room temperature for 30 min. The absorbance of extract was measured at 517 nm in UV spectrophotometer. Following formula was used to calculate the antioxidant activity (Rongshan *et al.*, 2012).

$$\frac{(A\ blank - A\ sample)}{A\ blank} \times 100 = \%\ DPPH\ scavenging\ activity$$

A blank represents the absorbance of the control reaction (containing all reagents except the tested compound) and A sample represents the absorbance of the tested compound. Ascorbic acid is used as a standard for antioxidant activity and results were expressed as % inhibition. The regression equation for this is y=0.392x+4.339 (R^2=0.999), all experiments were carried out in triplicate.

2.6 Antimicrobial studies

2.6.1 Antibacterial activity

The antibacterial activity of essential oil was determined using disc diffusion method as described by Bellik, (2014) with slight modification. Bacterial inocula was prepared in 10 mL of Müller-Hinton broth (Bioxon) and incubated at 37˚C for 24 hr. The inoculums were adjusted with sterile saline to obtain turbidity of the McFarland standard No. 0.5 (10^8 cfu/mL). Bacterial inocula were planted on Muller-Hinton agar plates, filter paper disc (5 mm diameter) impregnated with 100 µL of essential oil was placed on the plate. The plates were incubated at 37˚C for 24h. The inhibition zones were reported in mm. Three concentration of oleoresin (0.6, 0.8 and 1.0%) was used for this study.

2.6.2 Antifungal activity

The antifungal activity of essential oil was determined using disc diffusion method as described by Bellik, 2014 with slight modification. Fungal inocula were prepared in 10 mL of Müller-Hinton broth (Bioxon) and incubated at 37˚C for 24 hr. The inoculums were adjusted with sterile saline to obtain turbidity of the McFarland standard No. 0.5 (10^8 cfu/mL). Fungal inocula were planted on potato dextrose agar plates. On the surface of potato dextrose agar, impregnated (100 µl) filter paper discs (5 mm diameter) was placed.

The plates were incubated at 28˚C for 48 hr and the zone of inhibition was calculated, reported in mm. Three different concentration of oleoresin (0.6, 0.8 and 1.0%) were prepared in triplicate.

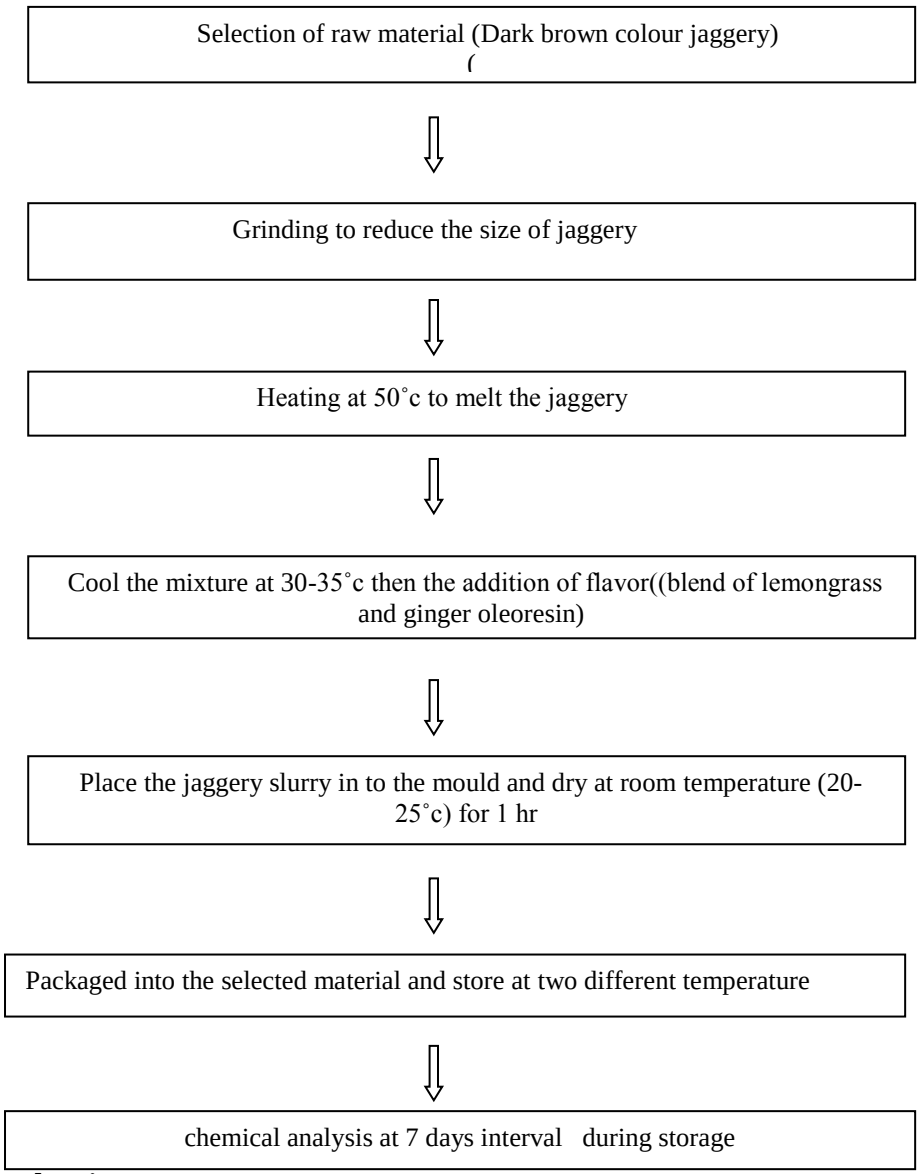
2.7 Formulation and optimization of product

After all prelimary analysis of oleoresign, three different concentration were selected initially to check the acceptability of concentration within the product. Further the cubes were analyzed for sensory evaluation and finalize the acceptable concentration of the raw ingredient.

Table 2.1 Formulation of flavor jaggery cubes

Ingredients	Control	J1	J2	J3
Jaggery (gm)	50	50	50	50
Oleoresin (BGL)(ml)	-	0.3	0.4	0.5

J0- Control
J1- Experimental sample (Jaggery 50 gm + oleoresin 0.3 ml)
J2- Experimental sample (Jaggery 50gm + oleoresin 0.4 ml)
J3- Experimental sample (Jaggery 50gm +oleoresin 0.5 ml)



2.8 Sensory evaluation

Sensory evaluation of flavor enriched jaggery cube was done by panel of semi trained judges on 9 point hedonic scale.

2.9 Storage study of flavor jaggery cubes

Optimized flavor sugar cubes was packed in two different packaging material [LDPE (low density polyethylene) and HDPE (high density polyethylene)] stored at two different temperature [(20-25˚) and elevated temperature (40-45˚)] to analyze the effect of external conditions on the samples. During storage, samples were analyzed for its bioactivity after every 7 days.

2.11 Statistical analysis:

Critical difference (CD) at 5% level of significance was recorded for comparison.

Result and conclusion

3.1 Formulation for flavor jaggery cubes by using blend of ginger and lemongrass oleoresin (BGL)

The study was carried out to optimize the concentration of BGL oleoresin for the better acceptability of the product. For this study different concentration of BGL were taken and analyzed for sensory evaluation. Semi trained panel of 10 members was developed for the study. The calculated results of obtained samples were reported in fig 3.1 According to the all panelists, sample J2 was selected because it has given the good taste and mouth feel to the product. From this study we can observed that 0.8 % concentration of J2 sample is acceptable, beyond this concentration (0.8%) strong flavour and mouth feel were observed which is not accepted by the panel. As mentioned above the reports of lemongrass oleoresin suggests that the high concentration of oleoresin provide strong aroma and taste due to the presence of aromatic compound i.e. citral (Baratta *et al*. 1998*;* Kasali *et al.* 2001; Nur Ain *et al*. 2011). Similarly, GO also contains bioactive compound which are responsible for taste and flavour . As a result, at highest concentration response of panelist has declined.

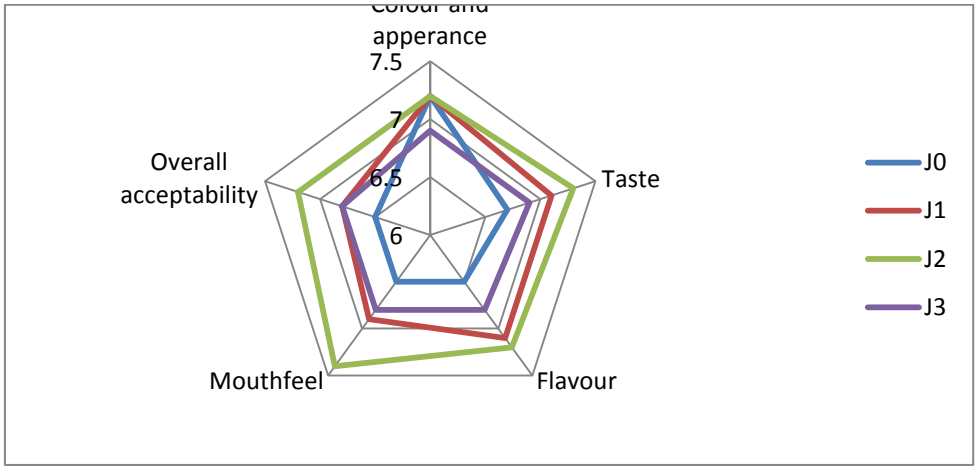


Fig 3.1 Organoleptic evaluation of BGL Jaggery cube

J0- Control

J1- Experimental sample (Jaggery 50 gm + oleoresin 0.3 ml)

J2- Experimental sample (Jaggery 50gm + oleoresin 0.4 ml)

J3- Experimental sample (Jaggery 50gm +oleoresin 0.5 ml)

3.2 Biochemical analysis of raw and finished product

Biochemical analysis of raw material (BGL oleoresin), BGL jaggery cube was carried out and presented in table3.1. The total phenolic content in BGL is 143.55mg/GAE/100gm, BGL jaggery cube is 104.23 mg /GAE/100 gm. Obtained results of oleoresin phenolic content are in agreement with Briones *et.al* (2014), Bellik et.al (2013) and Murad *et.al* (2011). The small change is due to the seasonal conditions and extraction procedures used for the experiment.

Antioxidant activity of samples was studied to check the efficacy of product against oxidation during storage. Scavenging activity of BGL oleoresin at optimized concentration (0.8%) is 50.72%,BGL jaggery cube is 39.41 %. Obtained results are in agreement with Elizabeth (2013) and Briones *et al*. (2015). The small change in antioxidant activity may be due to the presence of various types of compounds in them. Silva et al. (2000) also reported significant scavenging effects of phenolic compounds against the DPPH free radical. Hence, the presence of phenolic compounds such as eugenol, shogaols, zingerone, gingerdiols, gingerols in ginger oil responsible for their antioxidant properties. The solubility index of BGL jaggery cube was recorded as 1 min 30 sec.

Parameter	**BGL**	**BGL Jaggery cube**
Total phenolic content (mg/GAE/ 100gm)	143.55±0.17	104.23±0.12
Antioxidant activity (%)	50.72±0.24	39.41±0.11
Solubility index	-	1 min 30 sec

Values are represented as mean ± SD of three determinants

3.3 Antimicrobial activity of experimental samples

Three different concentrations of BGL were taken against gram positive and gram negative samples. BGL showed best activity against *B. Substilis* with increase in concentration. Maximum zone of inhibition were observed at higher concentration i.e. 1.0% ml followed by 0.8% and 0.6% ml shown in table 3.2 Therefore prepared BGL jaggery cube was analyzed at its optimized concentration (0.8%) and zone of respective samples was 7mm. Also the gram negative bacteria i.e. *E.Coli* was also shown activity with increase in concentration of BGL. The maximum zone of inhibition against *E.coli* was observed at higher concentration i.e 1.0% ml followed by 0.8% and 0.6% ml. As mentioned above jaggery cube was analyzed as its optimum concentration (0.8%) and it was 5mm. It can be seen that with increase in concentration of oleoresin antimicrobial activity increases while taste of product is decreases in terms on bitterness and provide burning sensation in throght. This may be due to the presence of phenolic compound i.e. **shogaols, zingerone and paradol** which are responsible for its activity. Antifungal activity of *A. niger* was found to be completely resistant towards the tested samples i.e oleoresin and BGL Jaggery cube. Obtained results are in agreement with Singh *et al.* (2008) and Bellik (2014). The antimicrobial activity of essential oils and oleoresins from spices and herbs is believed to be due to the phenolic compounds present in the oleoresin.

Table 3.2 Antimicrobial activity of raw and developed flavoured cubes

Microorganism	Zone of inhibition (mm) Oleoresin concentration			Flavour jaggery cube (0.4 ml)
	0.3 ml	0.4 ml	0.5 ml	
Bacteria				
B.substilis	7	10	12	7
E.Coli	6	8	10	5

3.4 Storage study of flavoured BGL jaggery cubes

Storage study of the developed samples was carried out at different factors to check the shelf life of the product at different storage condition. For this study two different storage temperatures [room temperature (20-25˚C) and elevated temperature (40-45˚C)] and two different packaging material (HDPE, LDPE) selected as a dependent variables for the responses. Sample were kept for 42 days and analyzed for its chemical activity

3.5 Chemical analysis of product

3.5.1 Effect of storage temperatures and Packaging material on phenolic content

The degradation curve is shown in graph fig 3.2, fig 3.3, fig 3.4 and fig 3.5 respectively The slight decrease in total phenolic content of BGL jaggery cubes was observed in both of temperature and packaging material during storage but the rate of degradation is less as compared to control This may be due to the oxidation of phenolic compounds and the stability of the compounds can be influence by the external factors like exposure to light, air, or different storage temperatures. Plant bioactive compounds are highly sensitive to the said factors as reported by **Moldovan *et.al* (2016).** Another logical reason behind this is volatility of oleoresin during storage which we have added into the sample. They are correlated with its activity as well.

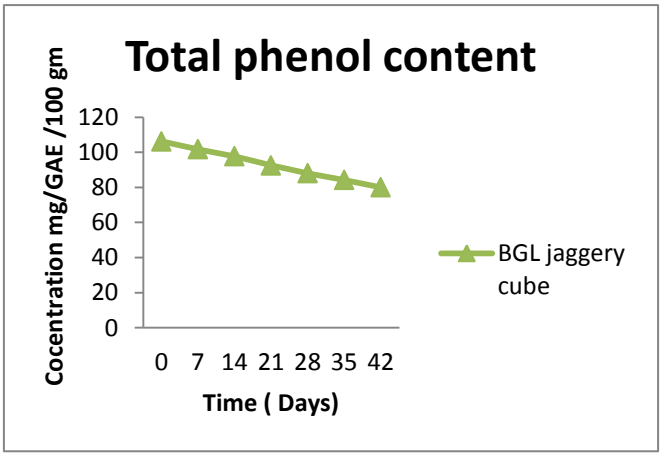


Fig 3.2 Effect of 20-25˚C temperature with HDPE packaging material on phenolic compounds

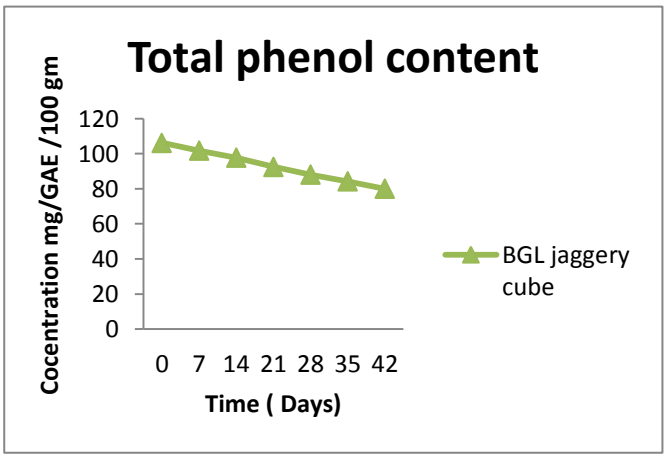
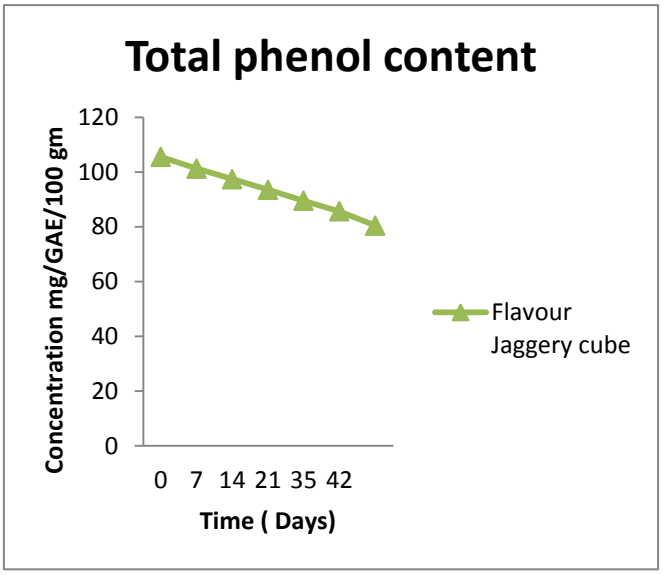


Fig 3.3 Effect of 20-25˚C temperature with LDPE packaging material on phenolic compound



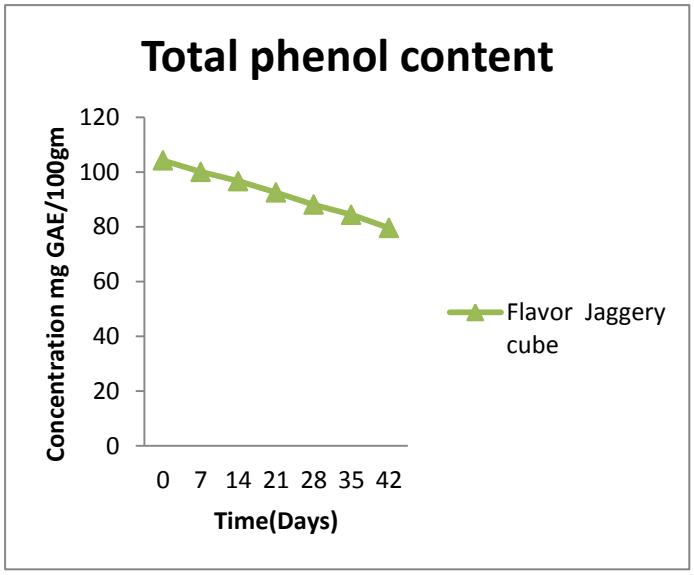
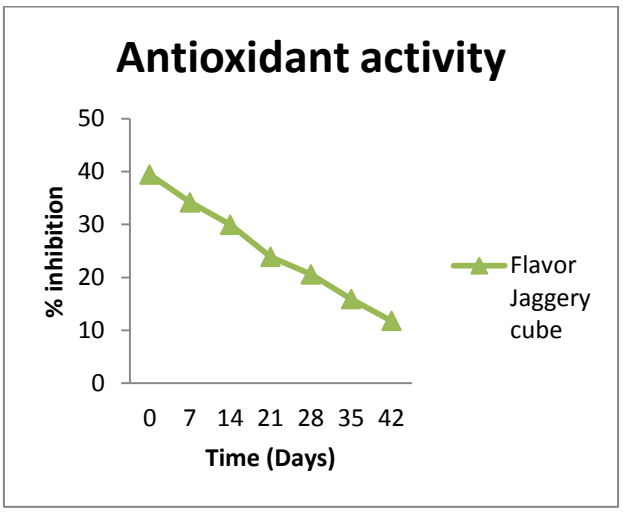


Fig 3.4 Effect of 40-45˚C with HDPE packaging material on phenolic compound

Fig 3.5 Effect of 40-45˚C with LDPE packaging material on phenolic compound

3.5.1 Effect of storage temperatures and Packaging material on antioxidant content

The changes were observed in antioxidant activity are reported in fig 3.6, fig 3.7, fig 3.8 and fig 3.9. it is observed that antioxidant activity of blended flavor jaggery cube was decreased with increase in time. Degradation rate of both the samples can be observe from the graph that slight degradation in antioxidant activity of BGL jaggery cubes was observed in both the packaging material during storage. As per the literature review, total phenolic content and antioxidant activity possesses positive correlation between each other. Degradation in antioxidant activity was observed due to the change in phenolic content with respective to the temperature.



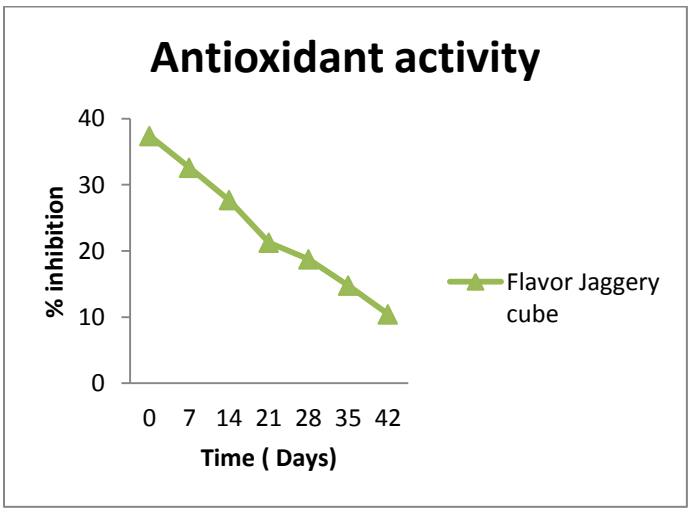


Fig 3.6 Effect of 20-25˚C temperature with HDPE packaging material on antioxidant activity

Fig 3.7 Effect of 20-25˚C temperature with LDPE packaging material on antioxidant activity

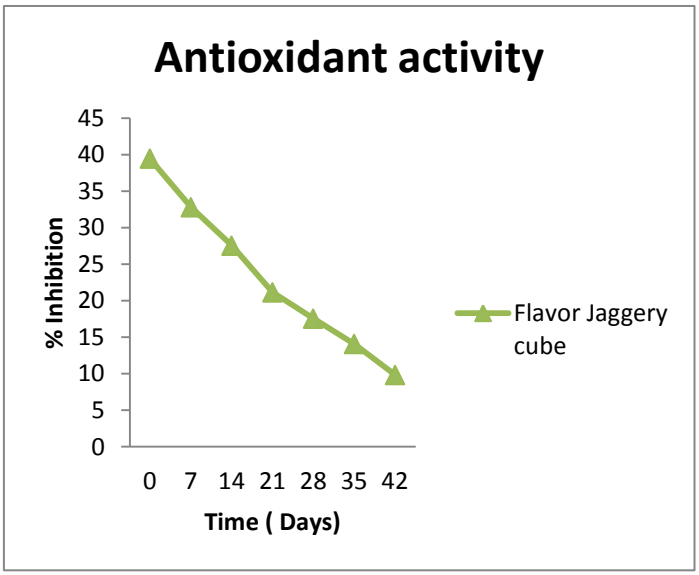


Fig 3.8 Effect of 40-45˚C temperature with HDPE packaging material on antioxidant activity

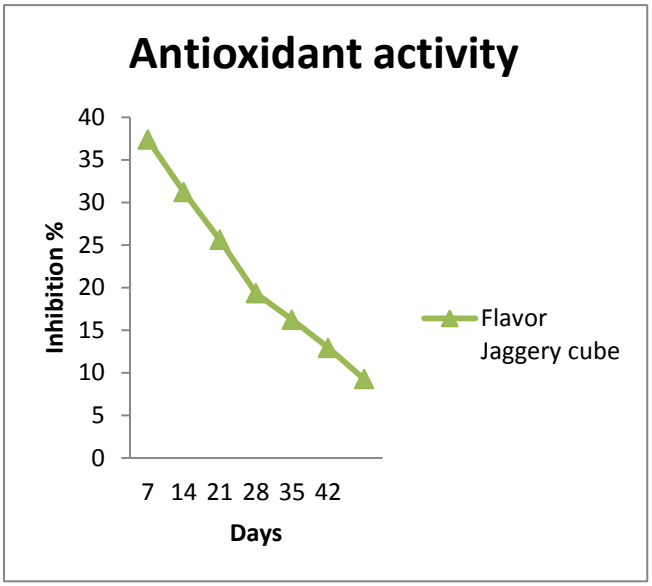


Fig 3.9 Effect of 40-45˚C temperature with LDPE packaging material on antioxidant activity

3.5.3 Effect of storage temperatures and Packaging material on moisture content

Effect of temperatures and packaging material on moisture content of BGL jaggery cubes was presented in figure 3.10,3.11,3.12 and 3.13. As evident from storage study, it can be seen that slight degradation in moisture content was observed at 0^{th} and 42 days respectively for both of temperature and packaging material This may due to the change in environmental conditions.

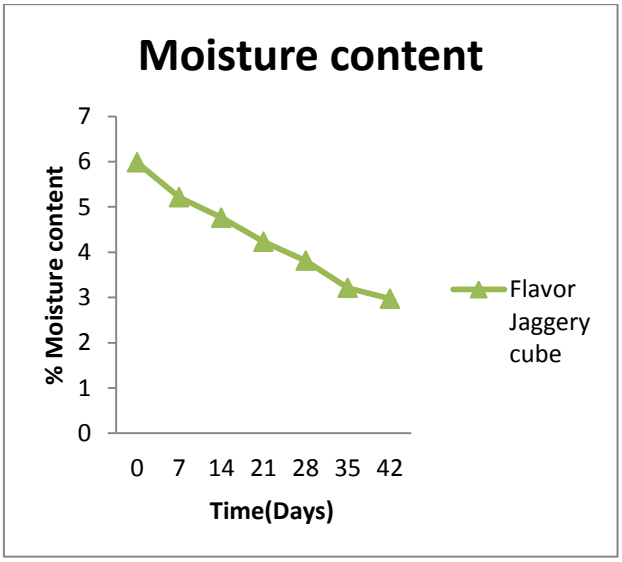


Fig 3.10 Effect of 20-25˚C temperature with HDPE packaging material on moisture content

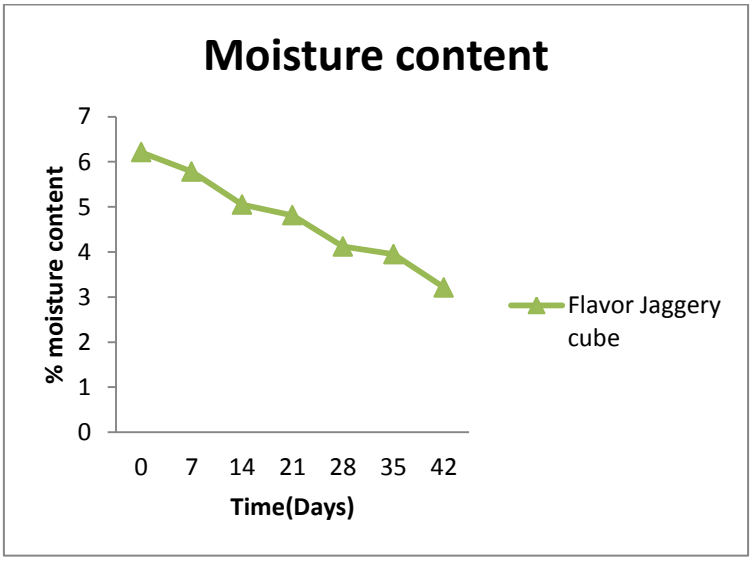


Fig 3.11 Effect of 20-25˚C temperature with LDPE packaging material on moisture content

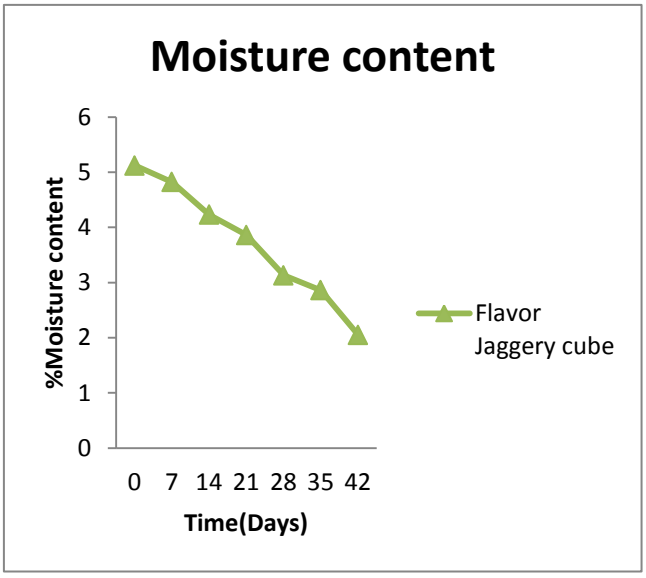


Fig 3.12 Effect of 40-45˚C temperature with HDPE packaging material on moisture content

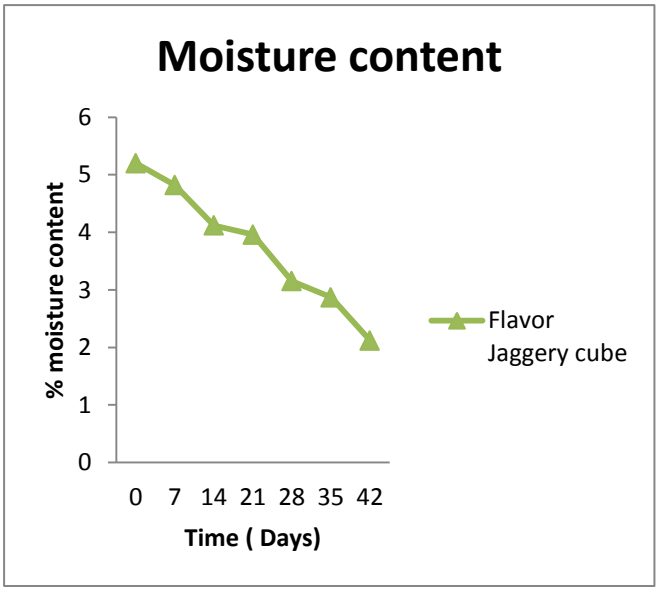


Fig 3.13 Effect of 40-45˚C temperature with LDPE packaging material on moisture content

SUMMARY AND CONCLUSION

The present investigation was carried out on "Development of flavour enriched jaggery cubes" as a source of instant energy to the beverages instead of sugar. The different concentration (0.6,0.8 and 1.0%) of raw material like blend of ginger lemongrass oleoresin was used for the flavor jaggery cube and as per the sensory evaluation score, 0.8% concentration of sample was selected for further antimicrobial and chemical studies. BGL and BGL jaggery cubes shows highest antimicrobial activity against *B. substilis* followed *E.coli* at optimized concentration (0.8%). During storage, bioactive compounds of flavor jaggery cubes and its antioxidant activity decreased slightly with respect to temperature and storage time.

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