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**Bhumendra Tripathi**  
Warner College of Dairy  
Technology, SHUATS,  
Prayagraj, Uttar Pradesh, India

**Rekha Rani**  
Warner College of Dairy  
Technology, SHUATS,  
Prayagraj, Uttar Pradesh, India

## Development of herbal lassi using giloy stem juice (*Tinospora cordifolia*)

**Bhumendra Tripathi and Rekha Rani**

### Abstract

In the present era of covid pandemic situation people are more health conscious, they like value added product as a part of their everyday meal or as well as the foods they eat for pleasure. India is one of the major countries in the world in regard to genetic resources of medicinal plants. Herbal lassi was prepared by using cow milk is a good and economical source of protein, vitamin, lactose, ash, are considered as a foodstuff of high nutritional value and one of the most affordable source of energy. The study was aimed and conducted to develop herbal lassi using giloy stem juice by utilizing cow milk, sugar and neem giloy stem juice. To adjudge the most acceptable levels of neem giloy stem juice in herbal lassi trials were conducted containing (3%, 6%, 9%). Among the different levels the best level was finalized on the basis of sensory and Physico-chemical analysis of product. The final optimized product contains 6% neem giloy stem juice was highly acceptable and can be used to develop a product without adversely affecting the sensory attributes of herbal lassi. The herbal lassi prepared without any addition of giloy juice is treated as control. The optimized product has fat (1.52%), moisture (81.10%), protein (3.086%), total solids (18.90%), ash (0.658%), carbohydrate (13.65%), acidity (0.518%), antioxidant (69.58%) and phytochemical content respectively in optimized herbal lassi. The yeast and mould was found 3.64 cfu/ml. The coliform count was absent in the product as per FSSAI standard.

**Keywords:** Herbal lassi, giloy stem, sugar, curd

### Introduction

Fermented milk products play an important role in human nutrition as far as therapeutic value of fermented milk is concerned. Importance of fermented milk in human nutrition has been well documented. Lassi is one of the popular, ready to serve indigenous fermented milk beverages. Lassi is served on very large scale in cold drink bars and restaurants during summer in almost every state in India. Many times "Chhash" is also referred to as "lassi" by technocrats in literature. However, Lassi is a popular product close to sweet stirred yoghurt has been used as a refreshing beverage from time immemorial in India, especially in western, northern and central regions. Lassi finds mention in ancient Indian scriptures along with its precursor dahi. Since ancient times, it has been used either with sugar (powdered sugar) or with black salt and zira. Along with all essential nutrients required for growth, development and tissue differentiation, fermented milks contain growth hormones – gastrin and insulin (Arora *et al.*, 2006). Lassi is also popular in some other parts of the world too, it is prepared by stirring whole curd into a delicious drink with addition of sugar or salt, a small amount of cold water or ice to make the product flowable. The popularity of this product is not only because of its refreshing and delicious taste, but also due to its nutritive and therapeutic benefits and thirst quenching quality. The health benefits of lassi are the result of biologically active components that are present in native milk and also, due to their suitably modulated activities produced through the action of lactic acid bacteria, recognition of the immense therapeutic and nutritional value and used for the treatment for diarrhea, dysentery, chronic specific and non-specific colitis, piles and jaundice. (Padghan *et al.* 2013) [5]. Several health benefits associated with the consumption of live probiotic bacteria are in controlling intestinal infections, improved digestion, improved lactose utilization, prevention of colon cancer, lowering of blood pressure, cholesterol, and reduced inflammation, etc. (Dugas *et al.* 1999).

Lassi is considered as digestive, nutritive and useful in gastrointestinal ailments. Lassi helps in digestion as it contains lactobacillus, which lubricates the intestines and smoothens the whole process of digestion. It is also an excellent source of probiotics (Times of India article-2019). *Tinospora cordifolia* (Giloy) is one of the important as well as very common herb which is normally used as a common ingredient in various Folk, Ayurvedic, Unani and Sridhar system of medicines. *Tinospora cordifolia* is a climbing shrub belongs to family Menispermaceae.

**Corresponding Author:**  
**Bhumendra Tripathi**  
Warner College of Dairy  
Technology, SHUATS,  
Prayagraj, Uttar Pradesh, India

It is commonly known as Guduchi, Amrita, Gurach, Tinospora. It is a large, glabrous deciduous climbing shrub. The stems are rather succulent with long filiform fleshy aerial roots form the branches. The bark is grey brown and watery. The leaves are membranous and cordate. The flowers are small and greenish yellow. This herb is found throughout tropical Asia. *Tinospora cordifolia* contains protein and rich in calcium and phosphorous. (Choudhary *et al.*, 2014)<sup>[1]</sup>.

Giloy has many aliphatic compounds and polysaccharides such as alkaloids, diterpenoid, lactones, glycosides, steroids, phenolic. All the parts of *Tinospora cordifolia* plant is used as therapeutic uses, it contains phytochemicals compounds. These compounds are used in cognition, anti-tumour, anti-inflammatory, anti-neoplastic, anti-osteoporotic, antimalarial, anti-hyperglycaemia, anti-hyperlipidaemia, antioxidant, anti-tuberculosis, anti-antigenic and anti-allergic. It is also used to control the side effects of cancer chemotherapy, radiotherapy and in surgery (Siddiqui *et al.*, 2013).

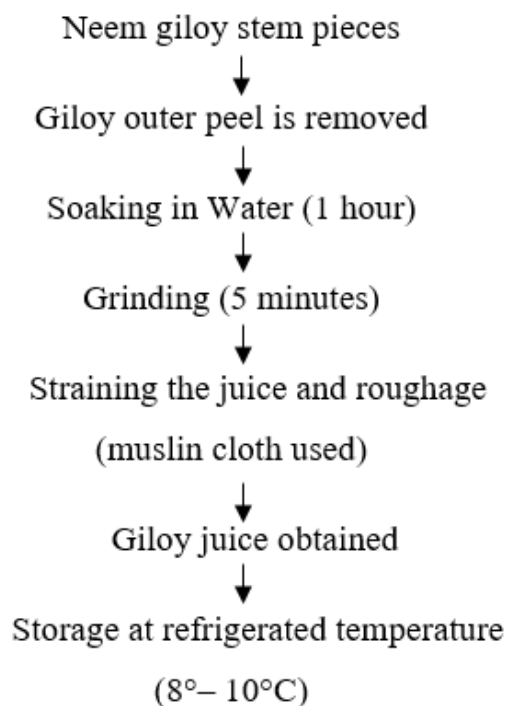
## Materials and Methods

### Collection of ingredients

Cow Milk and Sugar was collected from the local market at Prayagraj (U.P). Neem giloy stem juice is extracted by self in water as solvent and giloy stem is available in the local market. Starter Culture used of Essdee brand for curd formation and it is purchased from Essdee Marketing Pune (Type – XT), mixed strain of *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus*.

The study “Development of Herbal Lassi Using Giloy Stem Juice (*Tinospora cordifolia*)” was carried out in the laboratory of dairy technology, Warner College of Dairy Technology, Sam Higgin bottom University of Agriculture, technology and Sciences.

### Giloy stem juice extraction method



### Giloy juice stabilization

Only sugar syrup is added to remove the bitterness of the neem giloy juice no other process is done. Meanwhile there is a process of Ultra sonication used for reducing the bitterness

of giloy juice. (Sharma *et al.* 2020)



**Fig 3.2(a):** Neem giloy stem, (b) Chopped giloy stem, (c) Crushing giloy stem, (d) Soaked giloy stem, (e) Grinding giloy stem, (f) Giloy juice

### Treatment Combination (Ratio)

**T<sub>0</sub>:** Control sample normal lassi prepared from curd (100:00).

**T<sub>1</sub>:** Experimental herbal lassi sample prepared from curd and giloy juice (97:3).

**T<sub>2</sub>:** Experimental herbal lassi sample prepared from curd and giloy juice (94:6).

**T<sub>3</sub>:** Experimental herbal lassi sample prepared from curd and giloy juice (91:9).



**Fig 2:** Prepared sample treatments of herbal giloy lassi

### Preparation of Herbal Lassi

One liter of Cow milk was obtained and standardized to 3.0% fat and 8.5% solid not fat. The standardize milk is heated upto  $80\pm 2^\circ\text{C}$  for 10 minutes and the cooled upto  $37^\circ\text{C}$  before inoculation with starter culture @ of 1%. After inoculation the milk is incubated at  $40^\circ\text{C}$  for 6–7 hours. Then coagulum is broken and sugar syrup is added @ 15%, followed by giloy juice as per mentioned in the treatment to obtain the final product.

### Physico-Chemical analysis of developed herbal lassi

Determination of Total Solids– by Gravimetric method (FSSAI Manual) 2011. Determination of the Fat by Gerber method (IS: 1224-Part-1) (Reaffirmed 2012). Determination of Protein by (Kjeldahl method), (ISO8968-2:2001). Determination of As (IS:14433, Part 1) (Reaffirmed 2005). Determination of Titratable Acidity can (expressed as % lactic acid) BIS (IS: 1479, part: 1) 2016. Determination of Moisture content (AOAC-2000). Determination of the Carbohydrate (Lane Eynon) method in (FSSAI Manual- 2016). Determination of the Phytochemicals by (Screening test). Determination of Antioxidant (DPPH Method) (FSSAI Manual) 2016.

### Microbial Analysis

Yeast and Mould count IS: 5403(1999) (reaffirmed 2005) and coliform count IS: 5401-1 (2012) were determined by the following methods.

### Statistical Analysis

Data obtained is analyzed using Analysis of Variance (ANOVA) and Critical difference (C.D) in WASP software and excel software. The results obtained were statistically analyzed according to the method explained by Snedecor and Cochran (1994) [8]. The significance was evaluated on basis of critical difference.

### Phytochemical analysis screening test

Neem giloy stem was evaluated qualitatively for the presence of Alkaloids (Mayers's test), Glycosides (Borntrager test), saponins (Froth test), Flavonoids (Alkaline reagent test), Phenols (Liebermanns test), Terpenoids (Salkowski test), Tannins (Modified Prussian blue test) and Quinones (Acid test). The procedure for the analysis of these phytochemical constituents was adopted from Nandagoapalan, Doss, and Marimuthu (2016) [9] and Sharma (2020).

### Result and Discussion

The experimental finds of the study conducted have been tabulated and analyzed statistically using analysis of variance and critical difference technique. The significant and non-significant differences observed are analyzed critically within and between the treatment combinations. The results obtained are discussed below in the following sub heads:

#### Physico–chemical parameters Moisture content

The average value of Moisture percentage in control and experimental herbal lassi of different treatments are presented in Table 1. Statistically analyzed mean value of moisture of herbal lassi of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> was observed to be 80.24, 80.64, 81.10 and 81.57 respectively. The highest moisture content was observed in T<sub>3</sub> (81.57). Dhupal *et al.* (2018) [2] observed that the moisture content showed gradual increase in lassi with the increase in level of pudina extract. The treatment T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub> and T<sub>4</sub> were found to be 88.45, 88.68, 88.75 and 88.84. Thus the result reported by Dhupal is similar to herbal lassi.

#### Total Solid content

The mean value of Total Solid percentage in control and experimental herbal lassi of different treatments are presented in Table 1. Statistically analyzed mean value of total solid of herbal lassi of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> was observed to be 19.76, 19.36, 18.90 and 18.43 respectively. The highest total solid content was observed in T<sub>0</sub> (19.76). Kedaree *et al.* (2021) [4] reported that the total solid content showed gradual increase with the increase in level of kiwi pulp. This simultaneous increase from T<sub>0</sub> to T<sub>3</sub> may be due to high amount of total solid content of kiwi pulp (29.30) than cow milk (14.68) and curd (9.14). The lowest total solid content was noticed at T<sub>0</sub>. But in herbal lassi as the moisture content is increased the total solid is decreased.

**Table 1:** Average data obtained for different parameters of control and experiments of Development of Herbal Lassi using Giloy Stem Juice (*Tinospora cordifolia*)

Parameter Treatments	Score /value based on mean value of different parameter of treatment				C.D. value at 0.5%
	T0	T1	T2	T3	
<b>Physico-Chemical Analysis</b>					
Moisture (%)	80.24 <sup>a</sup> ±0.01	80.64 <sup>b</sup> ±0.01	81.10 <sup>c</sup> ±0.02	81.57 <sup>d</sup> ±0.01	0.035
Total solid (%)	19.76 <sup>a</sup> ±0.01	19.36 <sup>b</sup> ±0.01	18.90 <sup>c</sup> ±0.03	18.43 <sup>d</sup> ±0.01	0.040
Ash (%)	0.58 <sup>a</sup> ±0.01	0.59 <sup>b</sup> ±0.01	0.64 <sup>c</sup> ±0.01	0.65 <sup>d</sup> ±0.02	0.003
Protein (%)	3.22 <sup>a</sup> ±0.01	3.11 <sup>b</sup> ±0.01	3.08 <sup>c</sup> ±0.01	3.03 <sup>c</sup> ±0.02	0.035
Fat (%)	2.28 <sup>a</sup> ±0.07	1.80 <sup>b</sup> ±0.10	1.52 <sup>c</sup> ±0.11	1.12 <sup>d</sup> ±0.10	0.258
Total carbohydrates (%)	13.67 <sup>a</sup> ±0.07	13.84 <sup>a</sup> ±0.10	13.65 <sup>a</sup> ±0.10	13.62 <sup>a</sup> ±0.06	0.010
Acidity (%)	0.56 <sup>a</sup> ±0.03	0.54 <sup>ab</sup> ±0.01	0.51 <sup>ac</sup> ±0.01	0.47 <sup>ad</sup> ±0.01	0.050
Antioxidant (%)	37.58 <sup>a</sup> ±0.30	53.71 <sup>b</sup> ±0.70	69.58 <sup>c</sup> ±0.60	83.20 <sup>d</sup> ±0.70	1.840
<b>Microbial analysis</b>					
Yeast and mould (cfu/gm)	4.48 <sup>a</sup> ±0.05	3.93 <sup>b</sup> ±0.08	3.64 <sup>c</sup> ±0.05	3.42 <sup>d</sup> ±0.07	0.21
Coliform	Nil	Nil	Nil	Nil	Nil
<b>Sensory attributes</b>					
Colour and appearance	8.00 <sup>a</sup> ±0.30	6.80 <sup>a</sup> ±0.50	7.60 <sup>b</sup> ±0.37	6.00 <sup>a</sup> ±0.40	1.25
Flavour and taste	8.00 <sup>a</sup> ±0.30	7.00 <sup>a</sup> ±0.31	8.40 <sup>ab</sup> ±0.24	6.00 <sup>ac</sup> ±0.40	0.90
Consistency	7.20 <sup>a</sup> ±0.37	6.60 <sup>a</sup> ±0.50	8.20 <sup>a</sup> ±0.37	6.20 <sup>ab</sup> ±0.37	1.24
Overall acceptability	7.60 <sup>a</sup> ±0.24	6.80 <sup>ab</sup> ±0.37	8.00 <sup>ac</sup> ±0.30	6.00 <sup>bd</sup> ±0.40	1.17

The value expressed as Means ± SE (range) and values with different small letters superscripts row wise differ significantly at 5% level of significance (P≤0.05).

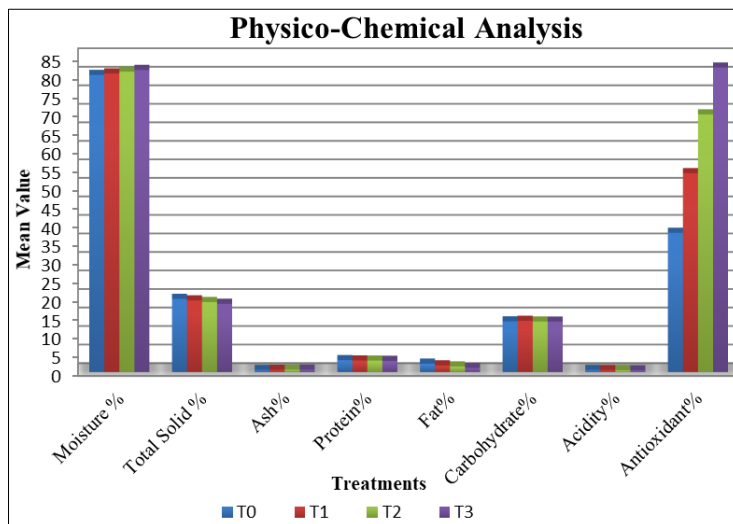


Fig 1: Average Physico-chemical analysis of control and experimental samples of Herbal lassi

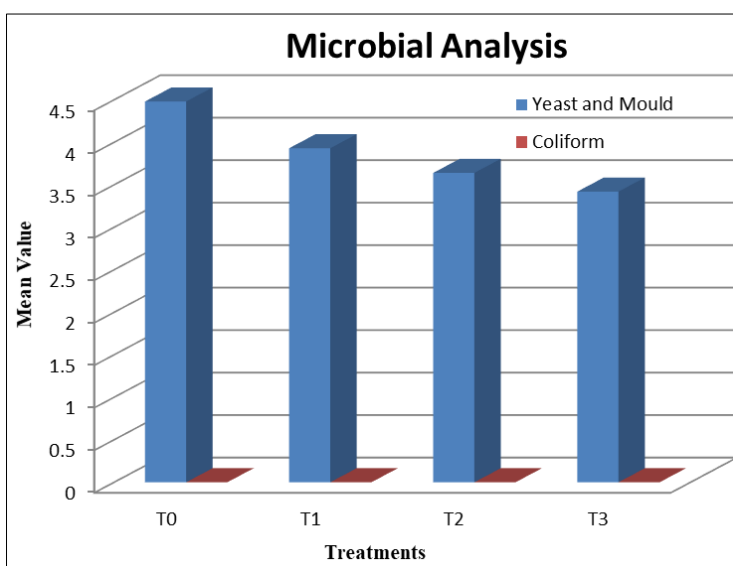


Fig 2: Average data on Yeast and Mould and Coliform of control and experimental sample of Herbal lassi

### Ash content

The mean value of Ash percentage in control and experimental herbal lassi of different treatments are presented in Table 1. Statistically analyzed mean value of ash of herbal lassi of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> was observed to be 0.585, 0.594, 0.642 and 0.658 respectively. The highest ash content was observed in T<sub>3</sub> (0.658). Upadhyay *et al.* (2017) [7] the ash content ranges from 0.68 to 0.81. Ash content increase as the carrot juice quantity is increased result obtained in herbal lassi is similar as reported by upadhyay. The ash content in the herbal lassi is increasing.

### Protein content

The mean value of Protein percentage in control and experimental herbal lassi of different treatments are presented in Table 1. Statistically analyzed mean value of Protein of herbal lassi of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> was observed to be 3.22, 3.11, 3.08 and 3.03 respectively. The highest protein content was observed in T<sub>0</sub> (3.22). Raut *et al.* (2015) [6]. The protein content of sample with different treatment combination was in range of 2.47 to 3.23 percent. The decrease in protein was there due to addition of varied proportion of mango pulp in yoghurt drink and as the protein content of the mango pulp is less than milk. In the herbal lassi the amount of dahi is

decreased according to the treatment therefore the protein is slightly decreasing.

### Fat content

The mean value of Fat percentage in control and experimental herbal lassi of different treatments are presented in Table 1. Statistically analyzed mean value of fat of herbal lassi of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> was observed to be 2.28, 1.80, 1.52 and 1.12 respectively. The highest fat content was observed in T<sub>0</sub> (2.28). Dhupal *et al.* (2018) [2] reported that higher fat content is observed in control treatment T<sub>1</sub>. The fat content of Lassi samples decreases as increase in the level of pudina extract. The treatments T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub> and T<sub>4</sub> were found to be 2.60, 2.53, 2.43 and 2.40. Thus the result reported similar to herbal lassi.

### Total Carbohydrate Content

The mean value of Total Carbohydrate percentage in control and experimental herbal lassi of different treatments are presented in Table 1. Statistically analyzed mean value of total carbohydrate of herbal lassi of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> was observed to be 13.67, 13.84, 13.65 and 13.62 respectively. The highest total carbohydrate content was observed in T<sub>1</sub> (13.84). Upadhyay *et al.* (2017) [7] stated that carbohydrate content ranges from 13.16 to 16.85. Carbohydrate content increases as increase in the level of carrot juice. In the herbal



lassi as the dahi level is decreased in the treatment the carbohydrate content start decreasing as carbohydrate present in giloy is not soluble in water.

### Acidity Content

The mean value of Acidity percentage in control and experimental herbal lassi of different treatments are presented in Table 1. Statistically analyzed mean value of acidity of herbal lassi of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> was observed to be 0.56, 0.54, 0.51 and 0.47 respectively. The highest acidity content was observed in T<sub>0</sub> (0.56). Kedaree *et al.* (2021) [4] reported that the average percent acidity of kiwi lassi were 0.86 (T<sub>0</sub>), 0.66 (T<sub>1</sub>), 0.60 (T<sub>2</sub>) and 0.54 (T<sub>3</sub>). It was observed that the acidity showed gradual decrease with the increase in level of kiwi pulp. Thus the result reported is similar to herbal lassi.

### Antioxidant Content

The mean value of Antioxidant percentage in control and experimental herbal lassi of different treatments are presented in table 1. Statistically analyzed mean value of antioxidant of herbal lassi of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> was observed to be 37.58, 53.71, 69.58 and 83.20 respectively. The highest antioxidant content was observed in T<sub>3</sub> (83.20). Upadhyay *et al.* (2017) [7] reported that Antioxidant ranges from 1.18 to 0.54. Decrease of antioxidant percent observed in the carrot lassi but in the Herbal lassi antioxidant percent is increasing with the increase in the giloy percent as giloy has more antioxidant activity compared to carrot.

### Yeast and mould Count

The mean value for yeast and mould counts ( $\times 10^3$ ) of Herbal lassi of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> was observed to 4.48, 3.93, 3.64 and 3.42 respectively and presented in Table 1. The herbal lassi was prepared by blending dahi with sugar and giloy juice. The highest recorded mean value was in T<sub>0</sub> (4.48) followed T<sub>1</sub> (3.93), T<sub>2</sub> (3.64) and T<sub>3</sub> (3.42). Data obtained in the Herbal lassi shows that as the percentage of the giloy juice increased in the treatments there is a decrease in the microbial growth i.e. giloy juice shows anti-microbial properties.

### Coliform Count

The mean value for coliform count was found nil in the product in all the treatments.

### Conclusion

The present research has shown that Giloy can be successfully used to prepare herbal lassi. Out of different treatments T<sub>2</sub> with 6% Giloy juice was found to be with the desired incorporated herbal lassi as per sensory attributes. Thus, it can be concluded that Giloy tried in this study could be successfully incorporated in herbal lassi without adversely affecting the sensory quality of the existing lassi. The lassi prepared was found to be safe on microbiological parameters and can be safely consumed. By adding giloy we are trying to increase the health benefit of the traditional beverage lassi and also the product is developed for the situation of covid pandemic. Ingredients like Giloy have beneficial role in cardio vascular diseases, natural antioxidants, which are effective in reducing the risk of heart disease, cancer, immune-system decline, cataracts and different inflammatory processes.

### Sensory Attributes Colour and Appearance

The mean value for colour and appearance of Herbal lassi of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> was observed to 8.0, 6.8, 7.6 and 6.0

respectively and are presented in Table 1. The data obtained in colour and appearance is judged by a panel of five judges using 9 point hedonic scale. The highest obtained score for colour and appearance was in T<sub>0</sub> (8.0) followed by T<sub>2</sub> (7.6), T<sub>1</sub> (6.8) and T<sub>3</sub> (6.0). The treatment T<sub>0</sub> has highest observed value.

### Flavour and Taste

The mean value for flavour and taste of Herbal lassi of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> was observed to 8.0, 7.0, 8.4 and 6.0 respectively and are presented in Table 1. The data obtained in flavour and taste is judged by a panel of five judges using 9 point hedonic scale. The highest obtained score for colour and appearance was in T<sub>0</sub> (8.0) followed by T<sub>2</sub> (8.4), T<sub>1</sub> (7.0) and T<sub>3</sub> (6.0). The treatment T<sub>2</sub> has highest observed value.

### Consistency

The mean value for consistency of Herbal lassi of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> was observed to 7.2, 6.6, 8.2 and 6.2 respectively and are presented in Table 1. The data obtained in consistency is judged by a panel of five judges using 9 point hedonic scale. The highest obtained score for consistency was in T<sub>2</sub> (8.2) followed by T<sub>0</sub> (7.2), T<sub>1</sub> (6.6) and T<sub>3</sub> (6.2). The treatment T<sub>2</sub> has highest observed value.

### Overall acceptability

The mean value for colour and appearance of Herbal lassi of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> was observed to 7.6, 6.8, 8.0 and 6.0 respectively and are presented in Table 1. The data obtained in colour and Appearance is judged by a panel of five judges using 9 point hedonic scale. The highest obtained score for colour and appearance was in T<sub>2</sub> (8.0) followed by T<sub>0</sub> (7.6), T<sub>1</sub> (6.8) and T<sub>3</sub> (6.0). The treatment T<sub>2</sub> has highest observed value.

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