EFFECT OF STORAGE TIME ON THE PHYSICOCHEMICAL PROPERTIES AND SENSORY ATTRIBUTES OF ALOE VERA READY-TO-SERVE (RTS) BEVERAGE

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Abstract
Purpose: Work was undertaken to process *Aloe vera* pulp into a ready-to-serve (RTS) beverage supplemented with mint (*Mentha requienii*) and ginger (*Zingiber officinale*) for their medicinal and nutraceutical value.
Design/methodology/approach: Incorporation of mint and ginger provides antiemetic and antioxidant properties to *Aloe vera* RTS. Physicochemical properties viz. pH, total soluble solids (TSS), titrable acidity, ascorbic acid, reducing sugar, total sugar, microbial load and sensory attributes of the beverage upon 60 days of storage (DOS) were studied and validation of results was done using statistical method. RTS was prepared using variable proportions of juice % and sugar in the beverage formulations viz. 10, 12, 14 and 16% respectively.
Findings: Beverage formulation containing 14% juice and 14% TSS was rated most acceptable by a panel of judges on a 9-point hedonic scale.

Keywords: *Aloe vera*, Mint (*Mentha requienii*), Ginger (*Zingiber officinale*), Physicochemical properties, RTS beverage

Paper type: Research paper

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INTRODUCTION

The true *Aloe vera* plant is called *Aloe barbadensis* Miller which belongs to the family Liliaceae. It is a perennial, drought resisting succulent plant. It has a history of use in folk medicine for treating skin and other disorders. The health benefits of *Aloe vera* have been propagated throughout the world. The bitter yellow and slightly sticky Aloe latex from the pericyclic tubules just beneath the outer skin of the leaves is commonly referred to as Aloe juice and it also has laxative properties. In the food industry, it is used as an ingredient for functional foods, mainly in the development of health drinks and beverages. Today, the Aloe industry is flourishing and the gel is used in many products, such as fresh gel, juice and other formulations for health, medical and cosmetic purposes (Enward and Benward, 2000). Proper scientific investigations on *Aloe vera* have gained more attention over the last decade due to its reputable medicinal, pharmaceutical and food properties. Photochemistry of *Aloe vera* gel has revealed the presence of more than 200 active substances including vitamins, minerals, enzymes, sugars, anthraquinones of phenolic compounds, lignin, saponins, sterols, amino acids and salicylic acid. Polysaccharides are considered to be the active ingredients for Aloe's anti-inflammation and immune modulation effects (Pugh *et al.*, 2001). Aloe leaf juice, dry leaf powder and gel are marketed in semi-processed forms and these products have great international demand (Agrawal, 1986).

Mentha arvensis is a genus of flowering plant from the family Lamiaceae. The leaf, fresh or dried, is the culinary source of mint. The leaves have a pleasant warm, fresh, aromatic, sweet flavour with a cool aftertaste. The substances which provide mint with its characteristic aroma and flavour are menthol and pulegone. Mint was originally used as a medicinal herb to treat stomach ache and chest pains.

Ginger is a tuber that is consumed whole as a delicacy, medicine or spice. It is the rhizome of the plant *Zingiber officinale*. Ginger contains up to 3% of a fragrant essential oil whose main constituents are sesquiterpenoids, with zingiberene as the main component. Smaller amounts of other sesquiterpenoids (β-sesquiphellandrene, bisabolene and farnesene) and a small monoterpenoid fraction (β-phelladrene, cineol, and citral) have also been identified. Coupled with increasing demand for soft drinks, there is considerable scope for developing naturally existing nutrient rich beverages from plants

like *Aloe vera*, mint and ginger. These beverages are far superior to synthetic beverages in terms of quality during the season. They are easily digestible, highly refreshing, thirst-quenching, appetizing and nutritionally far superior to many synthetic and aerated drinks (Srivastava and Kumar 2004). According to the Fruit Product Order (1955), the RTS beverage should have a juice content of not less than 10% (5% for lime), TSS of not less than 10% and preservatives as sulphur dioxide not more than 350 ppm or as benzoic acid not more than 600 ppm and 0.3% acidity. These beverages are not diluted before serving, hence the term ready-to-serve (RTS).

In the present study *Aloe vera* RTS beverage supplemented with mint and ginger was developed, since no previous work has been reported. The present study deals with the effect of storage time on physicochemical, microbial and sensory quality of *Aloe vera* RTS beverage and validation of results by using the STPR (social time preference rate) statistical method.

MATERIAL AND METHODS

Aloe vera leaves were procured from the Department of Botany, Banaras Hindu University, Varanasi. Leaves were sound, undamaged, mould/rot free and mature (3-4 years) in order to keep all the active ingredients in full concentration. Other raw materials including sugar, glass bottles, mint and ginger were procured from the local market. Apple and pineapple flavour essence and lemon yellow colour (Trishul brand) were also procured from the local market.

METHODOLOGY

Aloe vera pulp was taken out by the traditional hand filleting method to avoid contamination of internal fillet with the yellow sap. In this method, the lower 1 inch of the leaf base (the white part attached to the large rosette stem of the plant), the tapering point (2–4 inch) of the leaf top and the short, sharp spines located along the leaf margins were removed with a sharp knife, then the knife was introduced into the mucilage layer below the green rind, avoiding the vascular bundles, and the top rind was removed. The bottom rind was similarly removed and the rind parts, to which a significant amount of mucilage remained attached, were discarded. This is of critical concern because the highest

concentration of potentially beneficial Aloe constituents is found in this mucilage, as this layer represents the constituents synthesized by the vascular bundle cells empowered by energy developed in the green (chlorophyll-containing) rind cells through sun-induced photosynthesis. The filleting operation was completed within 36 hours of harvesting the leaves (Ramachandra and Srinivasa, 2008).

The pulp was heated to 60–65°C for 10 min. After heating, the pulp was mashed with the hand beater. The mashed pulp was strained with muslin cloth to retrieve the *Aloe vera* juice. The calculated amount of sugar was added and blended with juice to make four samples of 1200 ml each. The first sample (A_{10}) contained 10% juice and 10% TSS; the second sample (A_{12}) contained 12% juice and 12% TSS; the third sample (A_{14}) contained 14% juice and 14% TSS; and the fourth sample (A_{16}) contained 16% juice and 16% TSS. The juice contained *Aloe vera*, mint and ginger in the ratio of 80:10:10. The calculated amount of water was also added. Sodium benzoate and citric acid were added as preservative @ 0.2 % and 0.3 % per kg of pulp respectively. Citric acid was added to avoid a browning reaction, to improve the flavour of *Aloe vera* gel juice and to stabilize the juice. Apple flavour was added in the first and the third sample while pineapple flavour was added in the second and the fourth sample. The lemon yellow colour was also added in all four samples. RTS was centrifuged at 2000 rpm for 30 minutes at 25°C. After centrifugation, the RTS beverage was hot filled in clean and sterilized green bottles (200 ml) and sealed with crown corks with the help of a crown corking machine. Heat processing was then done by dipping in hot water (85–95 °C) for 1–3 minutes. The RTS bottles were then labelled and stored under refrigerated conditions (5°C) up to 60 days to determine the physicochemical, microbial and sensory quality of the beverage. Figure 1 represents the process flow chart for preparation of *Aloe vera* RTS.

PHYSICOCHEMICAL ANALYSIS OF *ALOE VERA* RTS

The pH values were determined with the help of an electronic pH meter (Thermo Scientific, 2 star), TSS measurement was done with the help of a hand refractometer (Bellingham Stanley Ltd., UK) (0-32 °C) and values were expressed as °Brix. Acidity of various samples was determined by titrating against 0.1 N NaOH according to AOAC (1995) method. Ascorbic acid content was determined by the titration method using 2,6-dichlorophenol endophenol dye ($C_{12}H_7NC_{12}$) as recommended

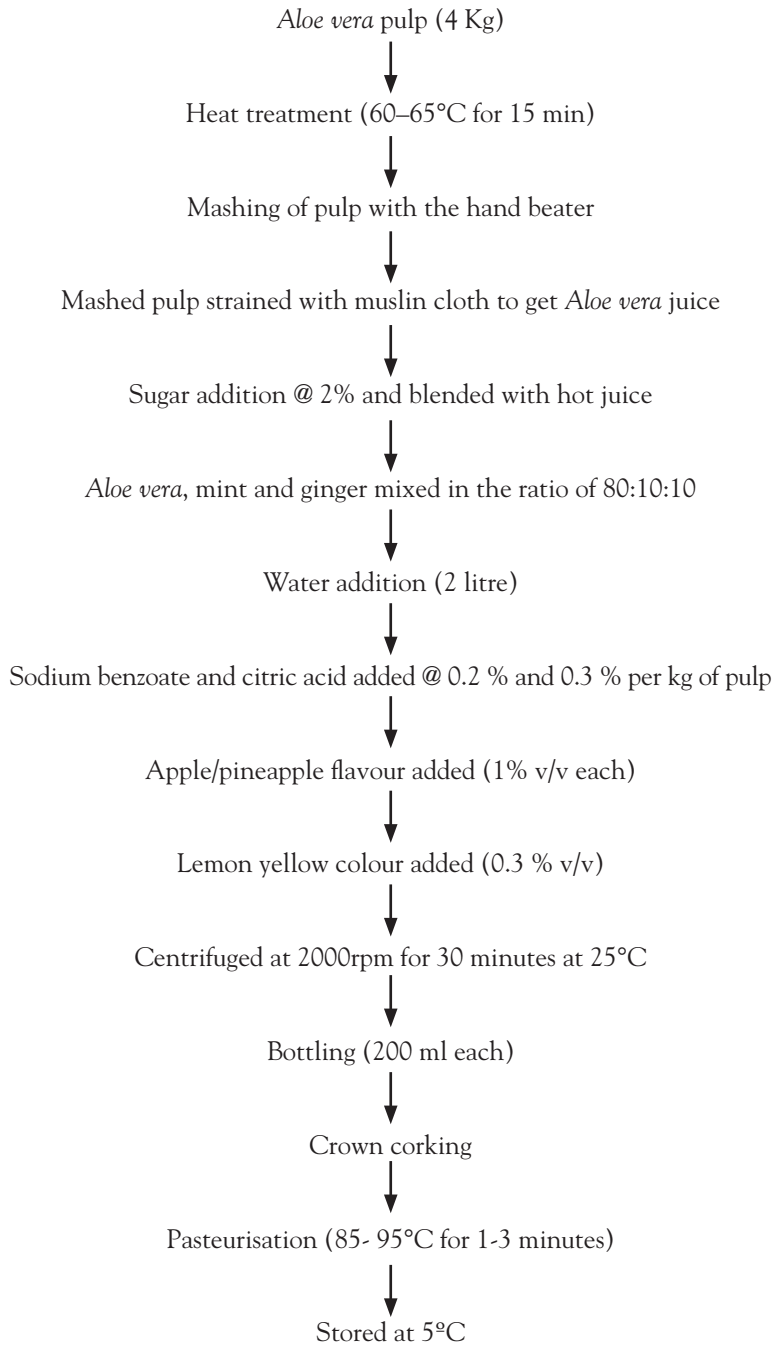

Fig. 1 The process flowchart for the preparation of the Aloe vera RTS

by Ranganna (2001). Reducing, non-reducing and total sugar were determined by the method recommended by Ranganna (2001).

MICROBIAL ANALYSIS

Microbial analysis was done to determine the total plate count (TPC) of the samples on the nutrient agar media for bacterial count by the method recommended by Harrigan and McCance (1966). Nutrient agar media was prepared and the samples were serially diluted up to 10^{-3} dilution factor. 0.25 ml of the samples, suspended in saline solution, were transferred to the respective petri dishes of nutrient agar media. Three replicates were taken for each dilution. The inoculated petri dishes were incubated in a BOD incubator for 48 hours at 37±1ºC and bacterial colonies were calculated by the following formula:

$$\text{TPC (cfu/ml)} = \text{No. of colonies} \times \text{dilution factor} / 0.25$$

DETERMINATION OF SENSORY QUALITIES

Sensory quality attributes viz. colour and appearance, consistency, flavour, taste and overall acceptability of the samples were evaluated using a 9-point hedonic rating test by a panel of six judges by the method recommended by Ranganna (2001).

STATISTICAL ANALYSIS

The data obtained from the various experiments were recorded and subjected to statistical analysis as per the Analysis of Variance method of Factorial Randomized Block Design (RBD). The significance and non-significance of data obtained from various experiments were judged with the help of an F (variance ratio) Table. The significant difference between the means was tested against the critical difference at the 1% and 5% level of significance by using STPR software for data analysis.

RESULTS AND DISCUSSION

Physicochemical characteristics of *Aloe vera* RTS

Aloe vera RTS physicochemical and sensory properties were studied for a period of 60 days. *Aloe vera* was supplemented with mint and ginger in

the ratio of 80:10:10 and the effects of storage time on pH, TSS, ascorbic acid and total sugar were studied.

Effect of storage period on physicochemical properties of *Aloe vera* RTS

Storage studies exposed the fact that pH decreased significantly during the storage period from 0 to 60 days (Fig 2 a). Freshly prepared *Aloe vera* RTS samples $A_{10,}$ $A_{12,}$ A_{14} and A_{16} exhibited pH values 3.23, 3.22, 3.22 and 3.23 respectively. Among the four samples, the minimum decrease in pH was observed in sample $A_{14,}$ which decreased from 3.22 to 3.16 after 60 days (Table 1). Similar trends were reported by Hamaran and Amutha (2007) in the case of banana and sapota beverage stored at different temperatures for 180 days. This gradual decrease in pH has a significant effect as lower pH does not allow pathogenic microorganisms to grow and hence acts as a preservative.

In the statistical analysis, it was found that the data are significant for different treatments and storage periods ($P \leq 0.01$). The combined effect of treatment and storage were found to be non-significant (Table 1). The interactive effect of mint extract and storage period had a significant effect on pH decrease due to the presence of flavenoids and terpenoids. Decrease in pH in all four samples may be due to the action of ascorbic acid on the sugar and protein component of RTS juice. Production of organic acids and amino acids leads to an increase in acidity and a decrease in pH as already reported for mango RTS (Kalra *et al.*, 1991; Sikder *et al.*, 2001).

TSS (Total Soluble Solid) significantly increased during the storage period from 0 to 60 for all four samples $A_{10,}$ $A_{12,}$ A_{14} and A_{16} (Figure 2 b). Maximum increase in TSS was observed in sample $A_{16,}$ which increased from 16.4 to 17.5 ºBrix, and minimum increase was observed in sample $A_{14,}$ which increased from 14.3 to 14.9 ºBrix. The increase in TSS during storage was also reported in RTS developed from guava nectar by Kalra *et al.* (1991) and Murari and Verma (1989). This increase in TSS might be due to conversion of insoluble polysaccharides into reducing sugars. The increase in reducing sugars can also be attributed to hydrolysis of sugars by acid, which might have resulted in degradation of disaccharides to monosaccharides. Another possible reason for the rapid increment in soluble solid contents may be due to hydrolysis of sucrose to invert sugars, as previously reported for increased TSS value with storage time in the case of bitter gourd RTS (Barwal *et al*, 2005).

Increased TSS concentration with storage period (of 60 days) is in correlation with the previous findings of Sirohi *et al.* (2005), who observed similar increased TSS in whey-based mango herbal mint beverage during storage, and attributed it to the solubilization of the insoluble portion of product due to the presence of acids (ascorbic and citric acid) in mint and ginger. Statistical analysis reveals that the data are significant for different treatments and storage period ($P \leq 0.01$) (Table 1). However the results of the present study in relation to TSS are in contradiction with those of TSS change observed in whey-based papaya RTS beverage, which observed no change in TSS during storage (Kumar and Manimegalai, 2005). This may be due to higher starch content in *Aloe vera* RTS supplemented with mint and ginger in comparison to papaya RTS, which undergoes hydrolysis in monosaccharide and other soluble sugars during storage.

From Fig 2 c, it is explicit that the titrable acidity significantly increased with storage time for all four samples ($A_{10,}$ $A_{12,}$ A_{14} and A_{16}). Among the four samples, the minimum increase in the acidity was observed in sample $A_{14,}$ which increased from 0.26 to 0.32%, and maximum increase in the acidity was observed in sample $A_{16,}$ which increased from 0.27 to 0.34 %. This can be attributed partly to the contribution of the inherent acid naturally present in the beverage and partly to the citric acid purposely added to the beverage at the time of preparation. Similar trends were reported by Hamaran and Amutha (2007), who observed an increment in acidity values of banana and sapota beverage stored at ambient conditions (35–36°C) and low temperature (3–5°C). Statistical analysis revealed that the combined effect of different treatments and storage period were significant ($P \leq 0.01$) (Table 1). The increase in acidity was observed in all treatments due to the formation of organic acids by ascorbic acid present in mint and ginger. The rise in acidity with increased storage time can be also attributed to degradation of polyphenol content in mentha extracts. Rapid conversion of proteins to amino acids is also responsible for increases in titrable acidity in *Aloe vera* RTS.

In pure *Aloe vera* juice, ascorbic acid content is less (5.6 mg/100 ml). Ginger and mint exhibit potential antioxidant and antiperoxidant properties due to the presence of certain polyphenols such as ascorbic acid, eugenol, rosmarinic acid and α-tocopherol (Rastogi and Mehrotra, 1995). The ascorbic acid content is higher in the case of mint (60 mg/100 g) and ginger (5mg/100 g), which improves the overall

Table I.
Physicochemical properties of *Aloe vera* RTS supplemented with mint and ginger

	Physicochemical properties						Sensory properties				
Sample	pH	TSS (°Brix)	Titratable acidity (%)	Ascorbic acid content (%)	Reducing Sugar (%)	Total sugar (%)	Colour and appearance	Flavour	Taste	Consistency	Overall acceptability
A_{10}	3.23	10.30	0.26	4.80	8.20	13.50	7.80	7.50	7.80	8.00	7.80
A_{12}	3.22	12.40	0.25	6.90	10.40	17.80	7.00	7.40	7.20	7.60	7.30
A_{14}	3.22	14.30	0.26	9.00	12.30	21.90	8.00	8.50	8.20	9.00	8.40
A_{16}	3.22	16.40	0.27	10.70	14.10	25.70	7.50	6.50	7.20	7.00	7.10

ascorbic acid content of *Aloe vera* RTS in comparison to pure *Aloe vera* juice. Incorporation of ginger prevents radiation-induced oxidative damage and also maintains the antioxidant properties of RTS due to the radioprotective action of eugenol present in ginger. However, the ascorbic acid content showed a decreasing trend with increased storage time (Fig 2 d). The ascorbic acid content in fresh *Aloe vera* RTS samples ($A_{10,}$ $A_{12,}$ A_{14} and A_{16}) was 4.80, 6.90, 9.00 and 10.70, respectively. After a storage period of 60 days, the final ascorbic acid content in respective samples ($A_{10,}$ $A_{12,}$ A_{14} and A_{16}) decreased to 4.00, 6.70, 8.90 and 10.30, respectively (Table 2). The minimum decrease was observed in sample $A_{14,}$ which decreased from 9.00 mg/ 100 ml to 8.40 mg/ 100 ml and the maximum decrease was observed in sample A_{16}, which decreased from 10.70 mg/ 100 ml to 9.70 mg/ 100 ml. The reduction in ascorbic acid during storage could be due to its oxidation to dehydro-ascorbic acid (Mokady *et al.*, 1997).

The degradation of ascorbic acid increased with storage time and this is attributed to an increase in moisture content with time. Increased moisture content leads to dilution of ascorbic acid and decreases its concentration. Moisture content in mint is higher (84.9 g/100 g), which may also be responsible for the decrease in the ascorbic acid content of *Aloe vera* RTS with storage time. Similar results were reported by Labuza *et al.* (1970) and Portenlanger and Heusinger, 1992, who proposed that rise in water activity (a_w) is responsible for the increased rate of ascorbic acid degradation. Atmospheric temperature, oxygen and the presence of trace metals also affects the ascorbic acid degradation during storage, as previously reported by Kirk *et al.* (1977).

A model system of dehydrated food was proposed by Kirk *et al.* (1977) which showed that the destruction of ascorbic acid was dependent upon water activity, moisture content and oxygen. The decrease in ascorbic acid was estimated from the first day of its preparation in *Aloe vera* RTS supplemented with mint and ginger in comparison to fresh *Aloe vera* juice due to its processing at high temperature (pasteurization). Ascorbic acid is a heat labile vitamin and during pasteurization, high temperature treatment will lead to its degradation.

Examination of the physicochemical properties of *Aloe vera* RTS suggests that the reducing sugar concentration increased significantly during the storage period of 60 days (Fig 2 e). Maximum increase was observed in samples A_{12} and $A_{16,}$ which increased from 10.4 to 11.2 %

and 14.1 to 14.9 %, respectively. Minimum increase was observed in sample $A_{14,}$ which increased from 12.3 % to 12.9 %. The increase in reducing sugar content in *Aloe vera* RTS may be due to hydrolysis of sucrose to glucose and fructose by the acid present in the beverages along with simultaneous decrease in the non-reducing sugars (Lotha, 1992). The combined effect of treatment and storage was found to be significant ($P \leq 0.05$) (Table 2). The increase in reducing sugar is due to inversion of non-reducing sugar to reducing sugars under acidic conditions, which correlates with earlier findings (Aruna *et al.*, 1997). Sethi (1982) also reported a similar trend in the case of lime ginger cocktail and jack fruit beverage. The increase in reducing sugars can be attributed to the hydrolysis of non-reducing sugars during processing and storage. The results of the present study contradict earlier reports by Barwal *et al.* (2005), who observed a decrease in sugar concentration in bitter gourd; however, in that trial, storage time was 90 days, which is much longer in comparison to the present study.

The total sugar content of freshly prepared *Aloe vera* RTS samples was 13.5, 17.8, 21.9 and 25.7 respectively. The storage period had a significant effect on total sugar content of *Aloe vera* RTS. From Table 2, it can be deduced that the total sugar content decreased significantly during the storage period of 60 days. Maximum decrease was observed in samples A_{12} and $A_{16,}$ which decreased from 17.8 to 17% and 25.7 to 24.9% respectively. Minimum decrease was observed in sample $A_{14,}$ which decreased from 21.9 to 21.3%. This minimum decrease in total sugars during storage may be attributed to accelerated hydrolysis of insoluble polysaccharides and other carbohydrate polymers (Narayana, *et al.*, 1996). The Mailard reaction and other chemical reactions of sugars with acids during storage also lead to a decrease in total sugar content. In the statistical analysis, it was found that the data are significant for different treatments and storage periods ($P \leq 0.01$) (Table 2).

Microbial analysis was performed at 30 days and 60 days of storage at 10^{-3} dilution. Maximum bacterial count was found in sample $A_{10,}$ which increased from 1.66×10^{-3} to 2×10^{-3}**.** Minimum bacterial contamination occurred in sample $A_{16,}$ which increased from 0.66×10^{-3} to 1×10^{-3}. Microbial load in the RTS beverages increased slightly with storage time. The microbial growth of RTS beverage decreased with increased sugar level because of the preservation properties of sugar. The physical and chemical results showed that, with respect to carbohydrate content, fruit pulp is an adequate

Table 2. Effect of storage period on pH, TSS and titratable acidity of *Aloe vera* RTS supplemented with mint and ginger

Storage periods	pH					TSS (°Brix)					Titratable acidity (%)				
	A_{10}	A_{12}	A_{14}	A_{16}	Mean	A_{10}	A_{12}	A_{14}	A_{16}	Mean	A_{10}	A_{12}	A_{14}	A_{16}	Mean
0	3.23	3.22	3.22	3.23	3.23	10.30	12.40	14.30	16.40	13.35	0.26	0.25	0.26	0.27	0.26
15	3.21	3.21	3.20	3.20	3.21	10.60	12.50	14.50	16.80	13.60	0.28	0.26	0.28	0.30	0.28
30	3.20	3.19	3.19	3.19	3.19	10.70	12.70	14.60	16.90	13.72	0.29	0.29	0.29	0.31	0.30
45	3.17	3.16	3.18	3.17	3.17	10.90	13.00	14.80	17.10	13.95	0.32	0.31	0.30	0.33	0.32
60	3.16	3.15	3.16	3.16	3.16	11.30	13.40	14.90	17.50	14.28	0.33	0.32	0.32	0.34	0.33
Mean for A	3.19	3.18	3.19	3.19	3.19	10.8	12.8	14.60	17.0	13.8	0.30	0.29	0.29	0.31	0.30
Effect			$CD_{(0.01)}$					$CD_{(0.01)}$					$CD_{(0.01)}$		
Treatment (A)			0.8461					0.8461					0.8461		
Storage (B)			0.9460					0.9460					0.9460		
A×B			0.1892					0.1892					0.1892		

substrate for the growth of bacteria. These results concur with those of Ukuku and Fett (2006). Chitra *et al.* (1998) reported the microbial population (bacterial count) of banana RTS beverage in the range of 1-12 x 10^6 and 1-6 x 10^6 cfu/ml for samples stored at room temperature and refrigerated conditions for up to 300 days of storage. Saravanan and Manimeglai (2000) reported the microbial load as 1-2 x 10^6 bacteria, 1-2 x 10^4 fungi and 1 x 10^5 per gram yeast in whey based papaya juice blended RTS beverage for up to 90 days of storage in refrigerated conditions, which was considered safe for consumption. Compounds reported to have antimicrobial activity include benzopyrans, xanthones, flavonoids and tannins (Ishiguro *et al.*, 1986). Mint and ginger are reported to exhibit antimicrobial and radioprotective properties. *Mentha* extract and mint oil have been shown to possess antibacterial and antifungal activities. Mentha also exhibits antimutagenic properties (the ability to enhance error-free repair of DNA damage) (Vokovic-Gacis and Simic, 1993). Leaves of certain varieties of mint such as *Mentha piperita* contain 7-O-rutinosides of eriodictyol and luteolin, while *Mentha* oil contains menthol, menthone, neomenthone, cineole, menthyl acetate, isomenthol, limonene and pinene (Rastogi and Mehrotra, 1995). Ginger is known to contain several antioxidant compounds such as gingerol, gingerdiol and shogaol, which possess antimicrobial activity against food spoilage organisms and seems to improve shelf life.

Effect of storage period on sensory attributes of *Aloe vera* RTS

Figure 3 represents the effect of storage period on sensory attributes viz; colour and appearance, taste, flavour, consistency and overall acceptability of *Aloe vera* RTS on a 9-point hedonic scale. The storage period significantly decreased the colour ratings of the beverage ($P< 0.05$) and the mean value of colour and appearance during a storage period of 60 days (Table 3). Initial score of colour and appearance in *Aloe vera* samples were 7.8, 7.0, 8.0 and 7.5, respectively. After 60 DOS, the final score for colour and appearance of all four samples decreased to 7.1, 6.1, 7.5 and 6.9 respectively. The maximum decrease was observed in sample $A_{12,}$ which decreased from 7 to 6.1. The minimum decrease was observed in sample $A_{14,}$ which decreased from 8 to 7.5. From Table 3, it is explicit that the score for flavour decreased significantly during the storage period of two months. Among the four samples, the minimum decrease was observed in sample $A_{14,}$ which decreased from 8.5 to 8.1. The decrease

Table 3. Effect of storage period on ascorbic acid content, reducing and total sugar of *Aloe vera* RTS supplemented with mint and ginger

Storage periods	Ascorbic acid content (%)					Reducing sugar (%)					Total sugar (%)				
	A_{10}	A_{12}	A_{14}	A_{16}	Mean	A_{10}	A_{12}	A_{14}	A_{16}	Mean	A_{10}	A_{12}	A_{14}	A_{16}	Mean
0	4.80	6.90	9.00	10.70	7.85	8.20	10.40	12.30	14.10	11.25	13.50	17.80	21.90	25.70	19.73
15	4.70	6.70	8.90	10.30	7.65	8.30	10.60	12.50	14.40	11.45	13.40	17.60	21.70	25.40	19.53
30	4.30	6.40	8.70	10.10	7.38	8.60	10.90	12.60	14.50	11.65	13.10	17.20	21.06	25.30	19.30
45	4.10	6.30	8.70	10.00	7.28	8.80	11.00	12.80	14.70	11.83	12.90	17.20	21.40	25.10	19.15
60	4.00	6.10	8.40	9.70	7.05	8.90	11.20	12.90	14.90	11.98	12.80	17.00	21.30	24.90	19.00
Mean for A	4.38	6.38	8.74	10.16	7.44	8.56	10.82	12.62	14.52	11.63	13.14	17.36	21.52	25.28	19.34
Effect															
Treatment (A)			$CD_{(.01)}$					$CD_{(.01)}$					$CD_{(.01)}$		
Storage (B)			0.1231					0.1216					1.4784		
A×B			0.1376					0.1360					1.6529		
			0.2752					0.2720					3.3058		

in flavour may be attributed to the loss of volatile aromatic substances (Thakur and Barwal, 1998).

The sensory score for taste also decreased significantly during the storage period of 60 days (Table 3). The minimum decrease was observed in sample $A_{14,}$ which decreased from 8.2 to 7.7. Maximum decrease was observed in samples A_{10} and $A_{16,}$ which decreased from 7.8 to 7.1 and 7.2 to 6.5 respectively. Reasons for the decrease in taste score may be attributed to the development of bitterness and increase in acidity in the samples during storage. Non-volatile phenylpropanoid-derived compounds, particularly gingerols and shogaols present in ginger may be responsible for the development of bitterness with increased storage time. Sensory score data clearly indicates that sample A_{14} was found to be best among the four samples. The sensory score for consistency also decreased significantly during the storage period of 60 days in all four samples. However the minimum decrease was observed in sample $A_{14,}$ which showed a decrease in sensory score from 9.0 to 8.4. Other samples showed more decrement in consistency with increased storage period. The maximum decrease in consistency was observed in sample $A_{12,}$ which decreased from 7.6 to 6.7 (Table 3). Similarly, the score for overall acceptability also decreased significantly during the storage period of 60 days. Sensory evaluation of RTS beverages revealed that the highest score for overall acceptability of 8.4 was recorded in sample $A_{14,}$ and the lowest of 7.1 in sample A_{16} at 0 day. As the storage period advanced, the treatments showed a reduction in the mean score from 8.4 at 0 day to 7.9 after 2 months of storage in sample $A_{14,}$ and from 7.1 to 6.4 in sample A_{16}.

Sensory score data for four samples reveals that sample A_{14} showed the least decrease in organoleptic quality with storage (Table 4).

Table 4. Changes in microbial load in Aloe vera RTS supplemented with mint and ginger during storage

	Storage period (Days)	
Samples	30 days	60 days
	10-3 dilution (CFU/ml)	10-3 dilution (CFU/ml)
A_{10}	1.66	2.00
A_{12}	1.33	2.00
A_{14}	1.00	1.66
A_{16}	0.66	1.00

Storage periods	Colour and appearance					Flavour					Taste				
	A_{10}	A_{12}	A_{14}	A_{16}	Mean	A_{10}	A_{12}	A_{14}	A_{16}	Mean	A_{10}	A_{12}	A_{14}	A_{16}	Mean
0	7.80	7.00	8.00	7.50	7.50	7.50	7.40	8.50	6.50	7.40	7.80	7.20	8.20	7.20	7.60
15	7.60	6.80	7.80	7.40	7.40	7.30	7.20	8.30	6.30	7.20	7.60	7.10	8.10	7.00	7.40
30	7.50	6.60	7.50	7.20	7.30	7.20	7.20	8.20	6.20	7.20	7.40	6.80	8.00	6.90	7.20
45	7.30	6.30	7.40	7.00	7.00	7.10	7.10	8.00	6.00	7.00	7.00	6.50	7.80	6.70	7.00
60	7.10	6.10	7.50	6.90	6.90	7.00	7.00	8.10	6.00	7.00	7.10	6.60	7.70	6.50	6.90
Mean for A	7.40	6.50	7.60	7.20	7.18	7.22	7.18	8.22	6.20	7.16	7.38	6.84	7.96	6.86	7.22
Effect			$CD_{(0.01)}$					$CD_{(0.01)}$					$CD_{(0.01)}$		
Treatment (A)			0.1231					1.1037					1.4784		
Storage (B)			0.1376					1.2340					1.6529		
A×B			0.2752					2.4680					3.3058		

Table 5. Effect of storage period on colour & appearance, flavour and taste of *Aloe vera* RTS supplemented with mint and ginger

	Consistency					Overall acceptability				
Storage periods	A_{10}	A_{12}	A_{14}	A_{16}	Mean	A_{10}	A_{12}	A_{14}	A_{16}	Mean
0	8.00	7.60	9.00	7.00	7.60	7.80	7.30	8.40	7.10	7.60
15	7.80	7.40	8.70	6.80	7.40	7.60	7.10	8.30	6.90	7.40
30	7.50	6.60	7.50	7.20	7.30	7.20	7.20	8.20	6.20	7.20
45	7.30	6.30	7.40	7.00	7.00	7.10	7.10	8.00	6.00	7.00
60	7.10	6.10	7.50	6.90	6.90	7.00	7.00	8.10	6.00	7.00
Mean for A	7.40	6.50	7.60	7.20	7.18	7.22	7.18	8.22	6.20	7.16
Effect			$CD_{(0.01)}$					$CD_{(0.01)}$		
Treatment (A)			0.1231					1.1037		
Storage (B)			0.1376					1.2340		
A×B			0.2752					2.4680		

Table 6. Effect of storage period on consistency and overall acceptability of *Aloe vera* RTS supplemented with mint and ginger

CONCLUSION

It was concluded that the sample with 14% juice content and 14% TSS supplemented with mint and ginger in a specified ratio (80:10:10) was found to be superior over the other samples ($A_{10,}$ $A_{12,}$ and A_{16}) in terms of physicochemical properties viz; pH, TSS, titratable acidity, ascorbic acid content, reducing and total sugar content, microbiological and organoleptic qualities. The developed RTS can be preserved at refrigeration temperature without adding any chemical preservatives with desirable consumer acceptability for up to 60 days. The product may serve as an excellent beverage owing to its functional and nutritional properties. It may also serve as a potential herbal beverage in the near future at a commercial scale.

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