Storage studies of pure strawberry juice preserved with sodium benzoate, potassium sorbate and anti oxidant stored at ambient temperature

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Abstract

The purpose of this research was to find out the influence of various sugar concentrartions and preservatives and keeping temperature i. e. ambient temperature (20-25°C) to study the physicochemical and sensory qualities of strawberry juice pres4rerved for twelve months. The samples were marked as, T_{A0}=Strawberry juice (7.5°brix) - no preservatives (control), T_{A1}=Strawberry juice (7.5°brix) with 0.1% sodium benzoate, T_{A2}=Strawberry juice (7.5°brix) with 0.1% potassium sorbate, T_{A3}=Strawberry juice (7.5°brix) with 0.05% sodium benzoate +0.05% potassium sorbate, T_{A12}=Strawberry juice (7.5°brix) - no preservatives (control), T_{A13}=Strawberry juice (7.5°brix) with 0.1% sodium benzoate, T_{A14}=Strawberry juice (7.5°brix) with 0.1% potassium sorbate, T_{A15}=Strawberry juice (7.5°brix) with 0.05% sodium benzoate +0.05% potassium sorbate. Highest increase was reported in Total Soluble Solids (TSS) content by T_{A1} and T_{A2} (20.00%) while lowest increase was reported in T_{A15} (9.33%) during storage. Maximum decline in pHrecorded in T_{A0} (17.94%) followed by T_{A12} (16.23%) while lowest decrease was reported in T_{A15} (8.33%). Maximum increase in titratable acidity reported in T_{A0} (167.40%) followed by T_{A2} (140.00%) while minimum increase reported in T_{A15} (102.38%). Maximum ascorbic acid decreased in T_{A0} (83.06%) while minimum decrease was recorded in T_{A15} (56.20%) followed by T_{A13} (58.73%). Maximum increase in reducing sugar content was reported in T_{A2} (19.65%) while minimum increase reported in T_{A15} (13.10%). Maximum decrease in non reducing sugar reported in T_{A0} (78.87%) followed by T_{A12} (70.42%) while minimum decrease reported in T_{A15} (11.26%). Decrease in color was observed for all samples. Flavor deterioration reported in control samples (TAO and TA12) while treated samples showed minimum loss in flavor during storage. Comparatively better consistency was observed by treated juice. After storage of ninety days, the samples were rejected on the basis of color, flavor, consistency and overall acceptability. Among all the treatments TAO and TA12 were rejected during storage while TA15 was found better than all other samples followed by T_{A13}and showed better storage stability than other samples.

Key words:Strawberry juice, benzoate, sorbate, sucrose, ambient temperature.

INTRODUCTION

Strawberry (Fragaria sp.) Is a member of herbaceous perennial of the "shifts" the families, Strawberry is one of the most important fruit among the berries and thus possess a very special place among small fruit plants (Sharma et al. 2009). It is believed that berries are effective againstvarious types of cancers (Navindra and P.Seeram. 2008). It is cultivated in many countries of the world, but is grown on large scale in the United States, Japan, Mexico, Italy, Lebanon (Sharma et al. 2009). Strawberries grown wildly with smaller size have a pleasant flavor (Ensminger et al. 2007). In Pakistan its cultivation and production is increasing continuously because of its scope and bright future for the farmers and higher returns in the economic point of view for the farmers. There are various factors which affect the productivity of the strawberry. The strawberries that are grown on scientific agronomic practices give higher production than conventional cultural cultivation (Tonture et al. 2009). Strawberries are grown in the tropical and subtropical regions. The fruits future in a short period of time and should be consumed soon after harvesting. Its transportation to far off places are difficult therefore extra care should be taken while its transportation. Strawberries are the first fruit of spring in the market. Due to the delicious flavor, color and structure, its demand is increasing in all parts of the world including Pakistan among the consumers. It is consumed as such and different products are prepared from it such as canned, cooked, sweetened, jam, jelly and frozen whole berries. Strawberries are used to prepare different products from it such as as purees, jam, juice and wine etc. (Sharma et al. 2009). The fruit is soft, red in color and sweet in taste. Glucose is the main sweetener of strawberries and about 50% of sugar is glucose in the strawberry. Main acids of this fruit are citric and some malic acids. The fruit is highly red due to the pigment anthocyanin which is responsible for red color. The flavor of the fruit is due to the volatile substances present in it. Strawberries are low caloric fruits and provide fiber but are a rich source of vitamin C than oranges. The components of strawberries are, vitamin C (64.0 mg), water (91.75 g), protein (0.61 g), fat (0.37 g) and carbohydrate (7.02 g), fiber (2.3 grams), calcium (14.0 mg) and potassium (166.0 mg/160 g), respectively, and vitamin A 27IU. Its pH is ranging from 3.27 -3.86, which helps in achieving stability of its color. Citric acid and malic acid are 0.58 to 1.35% that contributes to its excellent flavor. TSS is in the range 8.0 up to 11.5% which is ideal for its market value and juice extraction. Soluble solids / acid ratio from 8.52 to 13.79 range and is a good balance for the sweet tart flavor. With the passage of time the demand for the juice is increasing due to its excellent flavor and color. It is highly perishable fruit and a little fluctuation in the temperature can cause spoil the fruits so extra care should be taken.In a country like Pakistan, this fruit can't be preserved therefore this study will help to preserve this fruit in the form of juice preserved with different preservatives at different storage temperatures. This will reduce the load on market during its peak season and will help

the farmers to raise their lifestyle. Potassium sorbate is an important preservative for this juice (Ayub *et al.* 2010). Sodium benzoate can maintain the good quality of fruit juices during preservation (Ayub *et al.* 2010). It is believed that this study will enhance the beverage industry to prepare different products from this fruit to strengthen the economy of the country with healthy and natural juice and will provide the job opportunities to the people. This will provide healthy products to the consumers and will serve as a good source for foreign exchange.

MATERIALS AND METHODS

Healthy and fresh mature fruits were purchased from the local market and were brought to Food Processing and Analytical Laboratory of the Department of Food Science and Technology, The University of Agriculture Peshawar, Pakistan where research work was carried out. The strawberries were washed followed by sorting, the juice was extracted using juice extracting machine. The samples were numbered as, T_{A0} =Strawberry juice (7.5°brix) - no preservatives (control), T_{A1} =Strawberry juice (7.5°brix) with 0.1% sodium benzoate, T_{A2} =Strawberry juice (7.5°brix) with 0.05% sodium benzoate +0.05% potassium sorbate, T_{A12} =Strawberry juice (7.5°brix) - no preservatives (control), T_{A13} =Strawberry juice (7.5°brix) with 0.1% sodium benzoate, T_{A14} =Strawberry juice (7.5°brix) with 0.1% potassium sorbate, T_{A15} =Strawberry juice (7.5°brix) with 0.05% sodium benzoate +0.05% potassium sorbate.

Chemical analysis

The total soluble solids (TSS) were determined by using Abbe refractometer at ambient temperature (AOAC, 2000). Inolab digital pH meter was used for pH determination. Acidity was determined by dissolving a known weight of sample in distilled water and titration against 0.01 N NaOH using phenolphthalein as indicator (Srivastava and Sanjeev, 2003). Ascorbic acid was determined by the direct colorimetric method using 2, 6- dichlorophenol- indophenols as decolorizing agent by ascorbic acid in sample extract and in slandered ascorbic acid solution (AOAC, 2000). Reducing and non-reducing sucrose was determined by lane Eynon method (AOAC, 2000).

Sensory evaluation

A panel of ten judges selected from staff and students of food science department evaluated the product fortnightly for color, flavor, consistency and overall acceptability by the method of Larmond (1977) using a scale from 1 to 9, where 1 represented extremely disliked and 9 represent extremely liked.

Statistical analysis

The data obtained was subjected to statistical analysis using RCBD (Randomized Complete Block Design) and the means were compared by using LSD (Least Significant Difference) test (Steel and Torrie, 1980). For all the analyses, the alpha error was set at 0.05%.

RESULTS AND DISCUSSION

Chemical analysis

Our results indicated that storage period and temperature had significant effect on total soluble solids (TSS) during storage. The mean TSS values of all samples decreased from 7.50 to 7.08 during storage. Minimumincrease occurred in T_{A15} (9.33%)while maximum increase was observed in T_{A1} and T_{A2} (20.00%) during storage at ambient temperature. Decrease was observed in control samples (T_{A0} and T_{A12}) during storage. These results are in agreement with the results obtained by Zeb $et\ al.$ (2009) during preservation of grape juice stored at room temperature for one month preserved with sodium benzoate and potassium sorbate. Similar results were obtained by Hussain $et\ al.$ (2011) during storage of apple and apricot blended juice storage at refrigeration temperature for three months. Increase in total soluble solids may be due to break down of polysaccharides into monosaccharide and oligosaccharides while decrease may be due to fermentation of sugars into ethyl alcohol, carbon dioxide and water.

The mean pH values of all samples decreased from 3.49 to 3.10 during storage. During storage maximum decrease in pH content was observed in T_{A0} (17.94%) followed by T_{A12} (16.23%) while minimum decrease was observed in T_{A15} (8.33%) followed by T_{A14} (8.62%). Similar results were recorded by Mehmood *et al.* (2008) during study of the effect of pasteurization and chemical preservatives on the quality and shelf stability of apple juice stored at ambient temperature for three months. During storage of apple and apricot blended juices storage, preserved with sodium benzoate at refrigeration temperature for three months by Hussain *et al.* (2011), decrease in pH was recorded. Decrease in pH may be due to conversion of pectin into pectenic acid, which increases acidity and decreases pH of the juice.

The mean titratable acidity values of all samples increased from 1.51 to 3.42 during storage. Storage and treatments has a significant effect on the titratable acidity of strawberry juice. Maximum increase was observed in T_{A15} (102.38%) followed by T_{A14} (105.35%). Similarly increase in titratable acidity was observed by Zeb *et al.* (2009) during preservation of grape juice with sodium benzoate and potassium sorbate, stored at room temperature for one month storage period. The results of Ayub and Bilal (2001) are in agreement with our results, who observed an increase in acidity of pomegranate syrup, preserved under different light conditions and different packaging materials at room temperature for storage period of four months. This increase might be due to

acidic compounds formed by degradation or oxidation of reducing sugar and high temperature. Increase in titratable acidity may be due to the breakdown of pectin into pectenic acid or due to the formation of acid by the breakdown of polysaccharides or oxidation of reducing sugars.

Ascorbic acid is sensitive to heat, light and oxygen etc and is the most difficult vitamin to be preserved during storage. As it is the least stable vitamin, it decreases in the product during storage. The mean ascorbic acid values of all samples decreased from 35.40 to 11.36 during storage. Maximum decrease was observed in T_{A0} (83.06%) while minimum decrease was observed in T_{A15} (56.20%) followed by T_{A13} (58.73%). The results are in agreement with the findings of Zeb *et al.* (2009) during preservation of grape juice with sodium benzoate and potassium sorbate, stored at room temperature for one month storage period, who observed decrease in ascorbic acid content of grape juice. The results of Ayub and Bilal (2001) are in agreement with our results, who observed decrease in ascorbic acid content of pomegranate syrup, preserved under different light conditions and different packaging materials at room temperature for storage period of four months. The losses may be due to oxygen present in the product and headspace of the package or it may be due to high storage temperature.

Sugars are the most important constituent of fruit product and are essential factor for the flavor of the food product and also act as a natural food preservative. The mean reducing sugars values of all samples increased from 5.80 to 5.53 during storage. Results showed that reducing sugars increased during storage and maximum increase was observed in T_{A2} (19.65%) followed by T_{A1} (18.62%) while minimum increase was observed in T_{A15} (13.10%) followed by T_{A13} (13.79%) while control samples i. e. T_{A0} and T_{A12} showed decrease during storage. Mehmood $et\ al.$ (2008) observed an increase in reducing sugar of apple juice preserved with chemical preservatives stored at ambient temperature for threemonths. Increase in reducing sugar of apple and apricot blended juice, preserved with sodium benzoate at refrigeration temperature for three months, and was observed by Hussain $et\ al.$ (2011). This increase in reducing sugar might be due to conversion of sucrose to reducing sugars (glucose and fructose) primarily due to acids and higher temperature.

The mean non reducing sugars values of all samples decreased from 1.42 to 0.97 during storage. Maximum decrease in non reducing sugar was observed in T_{A0} (78.87%) followed by T_{A12} (70.42%) while minimum decrease was observed in T_{A15} (11.26%) followed by T_{A13} (14.08%). Mehmood *et al.* (2008) observed decrease in non reducing sugar of apple juice preserved with chemical preservatives stored at ambient temperature for three months. Increase in reducing sugar of apple and apricot blended juice, preserved with sodium benzoate at refrigeration temperature for three months, was observed by Hussain *et al.* (2011). This decrease in non reducing sugar might be due to conversion of sucrose to glucose and fructose, primarily due to increase in acidity and high storage temperature and storage period length.

Sensory analysis

The analysis of our data showed that storage period and treatments had a significant effect on sensory attributes (color, flavor, consistency and overall acceptability) of the strawberry juice. Loss in color was recorded in all treatments in which maximum loss was recorded in control samples. Off flavor was produced in control samples which may be due to fermentation process in which ethyl alcohol, carbon dioxide and water were produced. Anthocyanin is responsible for the red color of the product which is lost during storage at high temperature which in return protects the flavor of the juice so loss in color directly affects the flavor of the juice. Data showed that all the samples were rejected on the basis of sensory evaluation by the experts. The results are in agreement with the findings of Zeb et al. (2009) during preservation of grape juice with sodium benzoate and potassium sorbate, stored at room temperature for one month storage period, who observed decrease in color of grape juice. The results of Ayub and Bilal (2001) are in agreement with our results, who observed decrease in flavor of pomegranate syrup, preserved under different light conditions and different packaging materials at room temperature for storage period of four months. These results are in agreement with findings of Nilugen and Mahendran (2010), who observed decrease in consistency of ready to serve beverages prepared from palmyrah fruit pulp and stored at room temperature for six months.

Conclusion

From this research study it was concluded that sugar concentration, combination of preservatives and storage temperature play a positive role in extending the shelf life of strawberry juice. It was concluded that T_{A0} =Strawberry juice (7.5°brix) - no preservatives (control) and T_{A12} =Strawberry juice (7.5°brix) - no preservatives (control), were rejected while T_{A15} =Strawberry juice (20.5°brix) with 0.05% sodium benzoate +0.05% potassium sorbatemaintained better quality followed by T_{A13} =Strawberry juice (20.5°brix) with 0.1% sodium benzoate stored at ambient temperature for three months.

Table 1 Effect of chemical preservatives on TSS of strawberry juice stored at ambient temperature (20-25°C).

Treatment	Storage period in days							
	Initial	20	40	60	80	% Inc/ dec	Mean	
		l I	TSS (Total S	Soluble Solid	ls content of	juice	1	
T _{A0}	7.5	3.5	3.5	3.0	2.5	66.66 *	4.000 b	
T _{A1}	7.5	7.5	8.0	8.5	9.0	20.00	8.100 a	
T _{A2}	7.5	7.5	8.0	8.5	9.0	20.00	8.100 a	
T _{A3}	7.5	7.5	7.7	8.0	8.5	13.33	7.840 a	
T _{A12}	7.5	4.1	4.0	4.0	2.5	66.66 *	4.420 b	
T _{A13}	7.5	7.5	7.5	8.0	8.5	13.33	7.800 a	
T _{A14}	7.5	7.5	7.5	8.0	8.5	13.33	7.800 a	
T _{A15}	7.5	7.5	7.5	7.8	8.2	9.33	7.700 a	
Mean	7.500 a	6.575 a	6.713 a	6.975 a	7.088 a			

^{*} = decrease occurred in it.

Values followed by different letters are significantly ($p \le 0.05$) different from each other.

Table 2 Effect of chemical preservatives on pH of strawberry juice stored at ambient temperature (20-25°C).

Treatment	Storage period in days							
	Initial	20	40	60	80	% Decrease	Mean	
				pH va	lue			
T_{A0}	3.51	3.25	3.10	3.01	2.88	17.94	3.15 b	
T_{A1}	3.51	3.35	3.32	3.23	3.14	10.54	3.31 a	
T _{A2}	3.51	3.32	3.25	3.20	3.17	9.68	3.29 a	
T _{A3}	3.51	3.35	3.28	3.25	3.20	8.83	3.32 a	
T _{A12}	3.48	3.23	3.12	3.04	2.94	16.23	3.16 b	
T _{A13}	3.48	3.31	3.23	3.21	3.17	8.90	3.28 a	
T_{A14}	3.48	3.29	3.27	3.21	3.18	8.62	3.28 a	
T _{A15}	3.48	3.30	3.27	3.22	3.19	8.33	3.29 a	
Mean	3.49 a	3.30 b	3.23 c	3.17 d	3.10 e			

Values followed by different letters are significantly ($p \le 0.05$) different from each other.

Table 3 Effect of chemical preservatives on acidity (%) of strawberry juice stored at ambient

temperature (20-25°C).

Treatment	iperature (Storage period in days								
	Initial	20	40	60	80	% Increase	Mean			
			A	cidity (%) co	ontent of juic	ce				
T_{A0}	1.35	2.12	2.82	3.42	3.61	167.40	2.66 b			
T_{A1}	1.35	1.92	2.43	2.92	3.22	138.51	2.36 c			
T _{A2}	1.35	1.92	2.45	2.95	3.24	140.00	2.38 c			
T _{A3}	1.35	1.82	2.08	2.78	3.04	125.18	2.21 d			
T _{A12}	1.68	2.49	2.95	3.54	3.82	127.38	2.89 a			
T _{A13}	1.68	2.21	2.68	3.22	3.64	116.66	2.68 b			
T _{A14}	1.68	2.30	2.88	3.18	3.45	105.35	2.69 b			
T _{A15}	1.68	2.12	2.62	3.16	3.40	102.38	2.59 b			
Mean	1.51 e	2.11d	2.61 c	3.14 b	3.42 a					

^{* =} decrease occurred in it.

Values followed by different letters are significantly ($p \le 0.05$) different from each other.

Table 4 Effect of chemical preservatives on ascorbic acid of strawberry juice stored at ambient temperature (20-25°C).

Treatment		Storage period in days								
	Initial	20	40	60	80	% Decrease	Mean			
			Asco	rbic acid co	ntent mg/100) kg				
T_{A0}	31.30	20.11	10.53	5.52	5.30	83.06	14.55d			
T _{A1}	31.30	23.53	12.45	8.50	7.00	77.63	16.55cd			
T _{A2}	31.30	24.45	13.80	8.50	7.00	77.63	17.01c			
T _{A3}	31.30	26.94	15.70	10.50	8.50	72.84	18.59c			
T _{A12}	39.50	26.00	23.00	16.50	13.50	65.82	23.70b			
T _{A13}	3950	32.56	25.30	21.11	16.30	58.73	26.95a			
T _{A14}	39.50	32.19	24.11	20.95	16.00	59.49	26.55a			
T _{A15}	39.50	34.50	29.69	22.53	17.30	56.20	28.70a			
Mean	35.40a	27.53b	19.32c	14.26d	11.36e					

Values followed by different letters are significantly ($p \le 0.05$) different from each other.

Table 5 Effect of chemical preservatives on reducing sugar of strawberry juice stored at ambient

temperature (20-25°C).

Treatment	perature	Storage period in days								
	Initial	20	40	60	80	% Inc/dec	Mean			
				Reducing su	gar content					
T_{A0}	5.80	2.82	2.70	2.32	1.93	66.72*	3.11b			
T_{A1}	5.80	6.26	6.54	6.76	6.88	18.62	6.44a			
T_{A2}	5.80	6.28	6.56	6.80	6.94	19.65	6.47a			
T_{A3}	5.80	6.24	6.52	6.74	6.84	17.93	642a			
T _{A12}	5.80	3.20	3.09	2.98	1.93	66.72*	3.40b			
T _{A13}	5.80	6.12	6.34	6.48	6.60	13.79	6.26a			
T _{A14}	5.80	6.12	6.36	6.50	6.62	14.13	6.28a			
T _{A15}	5.80	6.10	6.32	6.46	6.56	13.10	6.24a			
Mean	5.80a	5.39a	5.55a	5.63a	5.53a					

^{*} = decrease occurred in it.

Values followed by different letters are significantly (p \leq 0.05) different from each other.

Table 6 Effect of chemical preservatives on non-reducing sugar of strawberry juice stored at ambient temperature (20-25°C).

Treatment		Storage period in days							
	Initial	20	40	60	80	% Decrease	Mean		
				Non-reducir	ng sugar				
T_{A0}	1.42	0.58	0.48	0.38	0.30	78.87	0.63 b		
T_{A1}	1.42	1.32	1.24	1.18	1.14	19.71	1.26 a		
T _{A2}	1.42	1.30	1.22	1.16	1.12	21.12	1.24 a		
T_{A3}	1.42	1.32	1.26	1.20	1.16	18.30	1.27 a		
T _{A12}	1.42	0.68	0.56	0.48	0.42	70.42	0.71 b		
T _{A13}	1.42	1.34	1.28	1.24	1.22	14.08	1.30 a		
T_{A14}	1.42	1.34	1.26	1.22	1.20	15.49	1.28 a		
T _{A15}	1.42	1.36	1.30	1.28	1.26	11.26	1.32 a		
Mean	1.42 a	1.15 b	1.07 bc	1.01 bc	0.97 с				

Values followed by different letters are significantly (p<0.05) different from each other.

Table 7 Effect of chemical preservatives on color of strawberry juice stored at ambient temperature (20-25°C).

Treatment	Storage period in days								
	Initial	20	40	60	80	% Decrease	Mean		
				Color sco	re rate				
T_{A0}	8.00	3.00	2.00	1.67	1.30	83.75	3.19 b		
T_{A1}	8.00	6.00	3.00	2.67	2.00	75.00	4.33 a		
T_{A2}	8.00	6.00	3.00	2.67	2.00	75.00	4.33 a		
T_{A3}	8.00	6.00	3.00	2.67	2.00	75.00	4.33 a		
T _{A12}	8.00	5.00	2.12	1.67	1.50	81.25	3.65 b		
T _{A13}	8.00	6.00	3.00	2.67	2.00	75.00	4.33 a		
T_{A14}	8.00	6.00	3.00	2.67	2.00	75.00	4.33 a		
T_{A15}	8.00	6.00	3.00	2.67	2.00	75.00	4.33 a		
Mean	8.00a	5.50b	2.765c	2.42c	1.85d				

Values followed by different letters are significantly ($p \le 0.05$) different from each other.

Table 8 Effect of chemical preservatives on flavor of strawberry juice stored at ambient temperature (20-25°C).

Treatment	Storage period in days							
	Initial	20	40	60	80	% Decrease	Mean	
				Flavor scor	e rate			
T_{A0}	8.00	1.67	1.50	1.30	1.30	83.75	2.75 b	
T_{A1}	8.00	5.30	2.30	2.00	1.67	79.12	3.85 a	
T_{A2}	8.00	5.30	2.30	2.00	1.67	79.12	3.85 a	
T _{A3}	8.00	5.30	2.30	2.00	1.67	79.12	3.85 a	
T _{A12}	8.00	1.67	1.50	1.30	1.30	83.75	2.75 b	
T _{A13}	8.00	5.30	3.30	2.67	2.00	75.00	4.25 a	
T _{A14}	8.00	5.30	3.30	2.67	2.00	75.00	4.25 a	
T _{A15}	8.00	5.30	3.30	2.00	2.00	75.00	4.12 a	
Mean	8.00a	4.39 b	2.47c	1.99 cd	1.70 d			

Values followed by different letters are significantly ($p \le 0.05$) different from each other.

Table 9 Effect of chemical preservatives on consistency of strawberry juice stored at ambient temperature (20-25°C)

Treatment	Storage period in days							
	Initial	20	40	60	80	% Decrease	Mean	
			C	Consistency s	score rate			
T_{A0}	7.00	5.50	2.00	2.00	1.30	81.42	3.56 b	
T_{A1}	7.00	6.00	2.67	2.00	1.30	81.42	3.79 ab	
T_{A2}	7.00	6.00	3.00	2.00	1.30	81.42	3.86 a	
T_{A3}	7.00	5.50	3.00	2.00	1.30	81.42	3.76 ab	
T _{A12}	7.00	5.50	2.00	2.00	1.30	81.42	3.56 b	
T_{A13}	7.00	5.70	3.00	2.00	1.30	81.42	3.80 ab	
T_{A14}	7.00	5.70	3.00	2.00	1.30	81.42	3.80 ab	
T_{A15}	7.00	6.00	3.00	2.00	1.30	81.42	3.86 a	
Mean	7.00a	5.738b	2.709c	2.00d	1.30e			

Values followed by different letters are significantly (p≤0.05) different from each other.

Table 10 Effect of chemical preservatives on overall acceptability of strawberry juice stored at ambient temperature (20-25°C).

Treatment	Storage period in days							
	Initial	20	40	60	80	% Decrease	Mean	
			Over	all acceptabi	lity score ra	te		
T_{A0}	8.00	2.60	2.00	1.67	1.30	83.75	3.11 b	
T_{A1}	8.00	6.00	2.30	2.30	1.30	81.25	3.98 a	
T_{A2}	8.00	6.00	2.30	2.67	1.30	81.25	4.05 a	
T_{A3}	8.00	6.00	2.30	2.67	1.30	81.25	4.05 a	
T _{A12}	8.00	2.60	2.00	1.67	1.30	83.75	3.11 b	
T _{A13}	8.00	5.30	3.00	2.30	2.00	81.25	4.02 a	
T _{A14}	8.00	5.70	3.00	2.30	2.00	81.25	4.10 a	
T_{A15}	8.00	5.70	3.00	2.67	2.00	81.25	4.14 a	
Mean	8.00a	4.98 b	2.48 c	2.28 c	1.37 d			

Values followed by different letters are significantly (p≤0.05) different from each other.

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