Journal of Pharmacognosy and Phytochemistry

Available online at www.phytojournal.com

E-ISSN: 2278-4136
P-ISSN: 2349-8234
JPP 2018; 7(1): 1796-1802
Received: 01-11-2017
Accepted: 02-12-2017

NS Thakur
Department of Food Science and Technology, Dr. YS Parmar University of Horticulture and Forestry, Nauni, Solan, HP, India

Nancy Thakur
Department of Food Science and Technology, Dr. YS Parmar University of Horticulture and Forestry, Nauni, Solan, HP, India

Hamid
Department of Food Science and Technology, Dr. YS Parmar University of Horticulture and Forestry, Nauni, Solan, HP, India

Pradeep Kumar
Department of Food Science and Technology, Dr. YS Parmar University of Horticulture and Forestry, Nauni, Solan, HP, India

Abhimanyu Thakur
Department of Food Science and Technology, Dr. YS Parmar University of Horticulture and Forestry, Nauni, Solan, HP, India

Correspondence
Hamid
Department of Food Science and Technology, Dr. YS Parmar University of Horticulture and Forestry, Nauni, Solan, HP, India

Formulation and optimization of vitamin C rich ready-to-serve juice based beverage from wild aonla fruits and its quality evaluation during storage

NS Thakur, Nancy Thakur, Hamid, Pradeep Kumar and Abhimanyu Thakur

Abstract
The present investigations were conducted to develop vitamin C rich RTS beverage from wild aonla (*Phyllanthus emblica* L.) fruits. Different treatment combinations of juice (10 %, 12 %, 14 % and 16 %) and TSS (12 °B and 15 °B) were tried to standardize proper combination for RTS beverage. Eight treatment combinations for the preparation of wild aonla drink were tried and drink recipe (T_7) with 14 per cent juice was found to be best on the basis of its some physico-chemical characteristics like TSS (15 °B), titratable acidity (0.42 %), pH (4.20), ascorbic acid (61.38 mg/100 g), total phenols (2.17 mg/g) and with highest sensory scores for colour (8.00), body (7.92), taste (8.10), aroma (7.90) and overall acceptability (7.95). Further standardized product was packed in PET (polyethylene terephthalate) and glass bottles under ambient (20-25 °C) and refrigerated temperature (4-7 °C) conditions for six months. Drink could be stored safely for a period of six months under both storage conditions and also in both packaging materials with minimum changes in chemical and sensory attributes. However, comparatively minimum changes in drink packed in glass bottle and stored under refrigerated storage conditions were observed as compared to PET bottle.

Keywords: wild aonla, vitamin C, RTS, quality evaluation, storage, antioxidant properties

Introduction
Wild aonla (*Phyllanthus emblica* L.) or Indian gooseberry belongs to family Euphorbiaceae and is indigenous to tropical South East Asia, particularly in Central and Southern India (Parmar and Kaushal, 1982) [1] from where it spread to Sri Lanka, Malaysia and China. It is widely distributed in Uttar Pradesh, Maharashtra, Gujarat, Rajasthan, Andhra Pradesh, Karnataka, Tamil Nadu, Haryana and Himachal Pradesh. Although the wild aonla is widely distributed in the forests of HP up to an elevation of 1450 m above mean sea level (Parmar and Kaushal, 1982) [1], but there are no records of its area and production. Aonla fruit is a rich source of ascorbic acid, phenols, sugars, pectin, starch and minerals (Nath *et al*., 1992) [2], besides fruit contains about 20 times more vitamin C than the citrus fruits. It is used in Ayurvedic and Unani systems of Indian medicines. This fruit is acrid, cooling, refrigerant, diuretic and laxative. It is useful in anaemia, artherosclerosis, cough, diarrhoea, dysentry, dyspepsia, haemorrhages, leucorrhoea and jaundice. It possesses antibacterial, anticarcinogenic, antiemetic, antioxidative, antipyretic, antitumour, antiviral, cardiotonic, expectorant activities (Singh and Singh, 2014) [3]. However, it is not consumed much as fresh fruit as it is highly acidic and astringent in taste. Therefore, it is necessary to convert the aonla juice into certain beverages before it can be consumed. But scattered information is available in the literature with respect to the development of value added products from wild aonla. As the nutritive value of fruit based beverages is much more than the synthetic products, which are available in the market throughout the country. If synthetic drinks can be substituted with the fruit juice, it would be beneficial to the consumers as well as fruit growers. Therefore, Keeping in view the availability of this fruit in the forests of HP and importance with respect to its quality characteristics, such as natural antioxidants especially vitamin C and phenols, the present studies were under taken with the objective to develop RTS beverage from this underutilized fruit and to study effect of storage on its chemical constituents.

Material and methods
The mature fruits of *Phyllanthus emblica* L. procured from Chandesh area of Mandi district of Himachal Pradesh in the year 2015-16 and used for various physico-chemical analysis and juice extraction.

The juice from the aonla fruits after washing with water and then shredding through grater was easily extracted by using hydraulic press machine. Fruit drink was prepared by mixing different proportions of fruit juice in different concentrations of sugar syrup as given in Table 1. Due to presence of higher amount of natural acid in fruit juice so no acid was added to drink. SO_2 (70 ppm) was added at the end of product preparation of drink packed in PET bottles whereas drink packed in glass bottles was heat processed.

Table 1: Treatment detail of fruit drink

Treatment	T_1	T_2	T_3	T_4	T_5	T_6	T_7	T_8
Juice (%)	10	12	14	16	10	12	14	16
TSS (ºB)	12	12	12	12	15	15	15	15

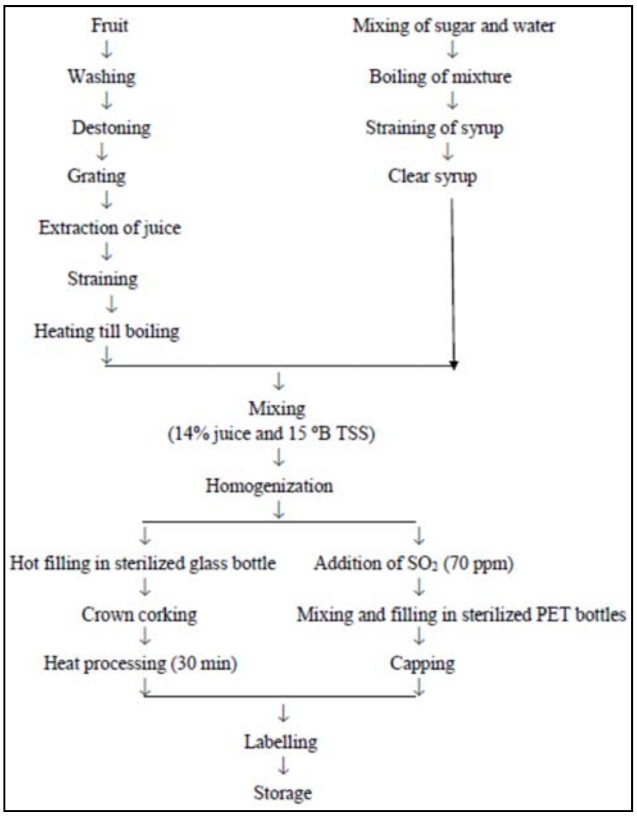

Fig 1: Unit operations for the preparation of wild aonla drink

Further standardized product was packed in pre-sterilised PET and glass bottles (200 ml capacity) (Fig. 1) under ambient (20-25 ºC) and refrigerated temperature (4-7 ºC) conditions for six months. The physico-chemical characteristics of the prepared product were estimated at zero, three and six months of storage. The colour of drink in terms of red (R) and Yellow (Y) units was observed with Tintometer (Lovibond Tintometer Model-E). The apparent viscosity of the drink was determined by using Ostwald viscometer and was expressed in time (flow rate in seconds) taken for samples to pass through the tube. TSS (ºB), titratable acidity (%), sugars (%) and ascorbic acid (mg/100 g) of prepared products were determined according to methods described by Ranganna (2009) [4]. The pH of the samples was determined by using a digital pH meter (CRISON Instrument, Ltd, Spain). Total phenols (mg/g) content was determined by Folin-Ciocalteu procedure given by Singleton and Rossi (1965) [5]. Nine points hedonic rating test was followed for conducting the sensory evaluation of drink as explained by Amerine *et al.* (1965) [6]. The panel of ten judges were selected to evaluate the product for sensory parameters such as colour, body, taste, aroma and overall acceptability. Data on physico-chemical characteristics of drink was analyzed by Completely Randomized Design (CRD) (Cochran and Cox, 1967) [7] before and during storage, whereas, data pertaining to the sensory evaluation were analyzed by using Randomized Block Design (RBD) as described by Mahony (1985) [8]. The experiments on recipe standardization and for storage studies were replicated three times.

Results and discussion

Standardization of recipe for the development of wild aonla drink

The data pertaining to physico-chemical and sensory characteristics of drink prepared by following different recipes are presented in Table 2 and 3.

Physico-chemical characteristics

Data presented in Table 2 indicate that maximum (0.33) red TCU were recorded in T_1 which were statistically at par with T_2 and lowest (0.17) in T_8 which was statistically at par with T_4 and T_7. The highest (2.69) yellow TCU were recorded in T_8 which was statistically at par with T_4 and T_7 while lowest (2.30) was found in recipe T_1 which was statistically at par with T_5.

Table 2: Physico-chemical characteristics of different recipes of wild aonla drink

Treatments	Physico-chemical characteristics						
	Colour (TCU)		TSS (ºB)	Titratable acidity (%)	pH	Ascorbic acid (mg/100 g)	Total phenols (mg/ g)
	R	Y					
T_1	0.33	2.30	12.00	0.32	4.31	50.79	1.48
T_2	0.28	2.47	12.00	0.36	4.24	56.92	1.81
T_3	0.25	2.56	12.00	0.42	4.20	61.42	2.17
T_4	0.20	2.67	12.00	0.49	4.15	67.92	2.43
T_5	0.30	2.36	15.00	0.32	4.31	50.78	1.47
T_6	0.26	2.50	15.00	0.36	4.24	56.89	1.80
T_7	0.22	2.62	15.00	0.42	4.20	61.38	2.17
T_8	0.17	2.69	15.00	0.49	4.14	67.86	2.42
$CD_{0.05}$	0.07	0.13	-	0.01	0.08	1.62	0.72

The highest content of titratable acidity (0.49 %) was recorded in T_4 and T_8 and the lowest (0.32) in recipe T_1 and T_5. The highest pH content (4.31) value was recorded in T_1 and T_5 and which was statistically at par with T_2 and T_6 the lowest (4.14) in recipe T_8 and which was statistically at par with T_3, T_4 and T_7. The ascorbic acid content of drink varied from 50.78 to 67.92 mg/100 g in the product and highest (67.92 mg/100 g) was recorded in T_4 which was statistically at par with T_8. The lowest (50.78mg/100 g) ascorbic acid content was recorded in T_5 which was statistically at par with T_1. Total phenols content was recorded highest (2.43 mg/ g) in T_4 which was statistically at par with T_2, T_3, T_6, T_7 and T_8 and lowest (1.47 mg/ g) in T_5 which was statistically at par with T_1, T_2, T_3, T_5, T_6 and T_7.

Data presented in Table 2 shows that recipe T_4 and T_8 contain higher content of titratable acidity, total phenols and ascorbic acid which are because of higher content of juice used in these recipes as compared to other recipes. The variation in juice content in different recipes have also contributed towards the variation in the colour units (yellow and red TCU) of the product. Our results are in conformity with earlier findings of Hamid *et al*. (2017) [9] in mulberry RTS beverage.

Sensory Characteristics

Data on sensory characteristics of different recipes of wild aonla drink given in Table 3 indicate that the highest (8.00) colour score was obtained in recipe T_7 while T_1 got the (7.03) lowest score which was statistically at par with T_2, T_3, T_4 and T_5. The recipe T_7 obtained maximum body score of 7.92 and minimum (5.47) in T_5 which was statistically at par with T_1. The highest (8.10) score of taste was also awarded to T_7 while T_1 got the lowest score of 5.00. The maximum (7.98) aroma score for aroma was obtained in recipe T_8 which was statistically at par with T_4, T_6, T_7 and T_8. The highest (7.95) score of overall acceptability was awarded to recipe T_7 and the lowest to T_1 which was statistically at par with T_2 and T_5.

Table 3: Sensory characteristics (scores) of different recipes of wild aonla drink

Treatment	Colour	Body	Taste	Aroma	Overall acceptability
T_1	7.03	5.50	5.00	5.61	6.00
T_2	7.16	6.45	5.50	6.50	6.31
T_3	7.26	6.75	7.74	6.83	6.82
T_4	7.15	6.32	6.45	7.95	6.94
T_5	7.23	5.47	6.60	6.50	6.36
T_6	7.60	7.53	7.17	7.83	7.16
T_7	8.00	7.92	8.10	7.90	7.95
T_8	7.45	7.32	6.34	7.98	7.20
$CD_{0.05}$	0.31	0.27	0.22	0.42	0.49

The maximum scores for sensory parameters like colour, body, taste and overall acceptability obtained in recipe T_7 might be due to the higher juice content, best combination of juice and syrup, best sugar-acid blend in the product and finally all these factors might have led the judges to award the highest scores to this recipe. The higher aroma scores obtained in T_7 and T_8 might also be due to the higher juice content in these recipes. Nearly similar results have been reported by Hamid *et al*. (2017) [9] in mulberry RTS beverage.

Physico-chemical and sensory characteristics of standardized recipe of wild aonla drink

For the development of wild aonla drink (Table 2) a recipe (T_7) with 14 per cent juice, 15 ^{o}B and 0.42 per cent acidity was found to be best on the basis of its highest sensory characteristics scores viz. colour, body, taste, aroma and overall acceptability, and comparable physico-chemical characteristics like colour, titratable acidity, pH, total phenols and ascorbic acids. Higher comparable values of various physico-chemical characteristics of this recipe are because of the higher content of juice in this recipe as compared to others. However, the highest scores of sensory parameters of this recipe shows its superiority over others because of higher juice content, best combination of juice and syrup, best sugar-acid blend in the product. All these factors might have lead the judges to award highest sensory scores to this recipe.

Effect of storage on physico-chemical characteristics of wild aonla drink

Colour and apparent viscosity

There was a non-significant decrease in red and yellow TCU of wild aonla drink during storage. The overall effect of storage period on the red and yellow TCU of drink indicates that they decreased from 0.22 to 0.11 and 2.62 to 1.90. However, overall effect of storage conditions shows that (2.40) yellow TCU of drink were retained more in refrigerated storage conditions as compared to ambient storage conditions (2.11). The reason for decrease in colour units of drink during storage might also be due to browning caused by copolymerization of organic acids in the product. Minimum decrease observed in colour units of drink packed in glass bottle than PET bottle might be because of slower rate of chemical reactions in the product packed in glass bottle as a result of difference in their thermal conductance properties. Similar decreasing trend of red and yellow colour units in mulberry drink during storage has been reported by Hamid *et al*. (2017) [9]. Whereas, apparent viscosity of wild aonla drink in terms of flow rate increased significantly during the storage period (Fig. 2) which might be due to increase in TSS and soluble sugars which increased strain and shearing rate and decreased the flow index of the product. As the flow index decreases it helps to develop pseudo plasticity and increased the apparent viscosity (Bal *et al.*, 2014) [10]. Similar results have been reported by Thakur *et al.* (2017) [11] in box myrtle drink.

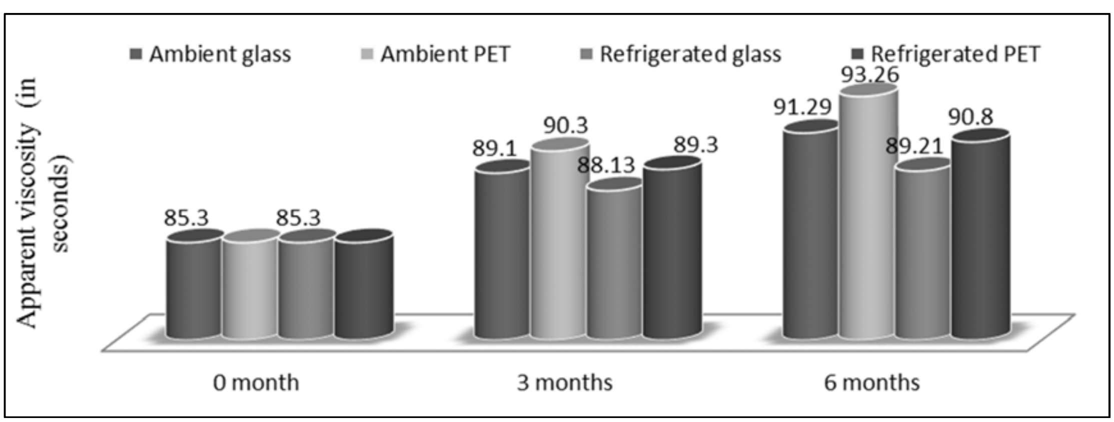

Fig 2: Effect of packaging and storage on apparent viscosity of wild aonla drink

TSS and sugars

Slight increase was experienced in TSS of wild aonla drink during storage (Fig. 3) might be due to the hydrolysis of polysaccharides into monosaccharide and soluble disaccharides (Gould, 1983) [12]. These results are also in conformity with the findings of Thakur *et al.* (2011) [13] in wild pomegranate drink during storage. During storage there was a gradual increase in reducing sugars and total sugars of wild aonla drink. Increase in sugars during storage might be attributed due to the hydrolysis of starch into sugars (Heikal *et al.*, 1964)[14]. However, more increase in reducing sugars (Fig. 4) was found in drink stored under ambient conditions as compared to refrigerated storage conditions due to the faster rate of reactions because of high temperature in ambient conditions. Similar trend of increase in sugars have been reported by Kumar *et al.* (2010) [15] in aonla drink. As far as the packaging material is concerned, more increase in sugars recorded in drink packed in PET bottle as compared to glass bottle might be due to faster rate of chemical reactions in the product packed in PET bottle as a result of their thermal conductance properties. Similar results have been reported by Thakur *et al.* (2011) [13] in wild pomegranate drink and Hamid *et al.* (2017) [9] in mulberry RTS beverage. However, the increase in total sugars was lower in refrigerated storage conditions (increased from 11.55 to 11.83 and 11.92 % in both glass and PET) than ambient conditions (increased from 11.55 to 12.10 and 12.21 % in both glass and PET).

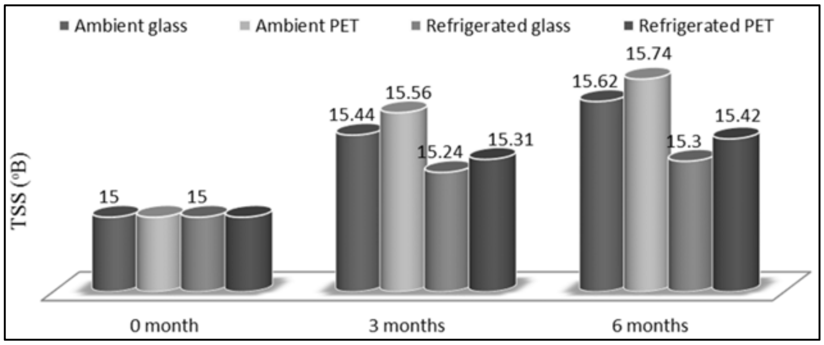

Fig 3: Effect of packaging and storage on TSS of wild aonla drink

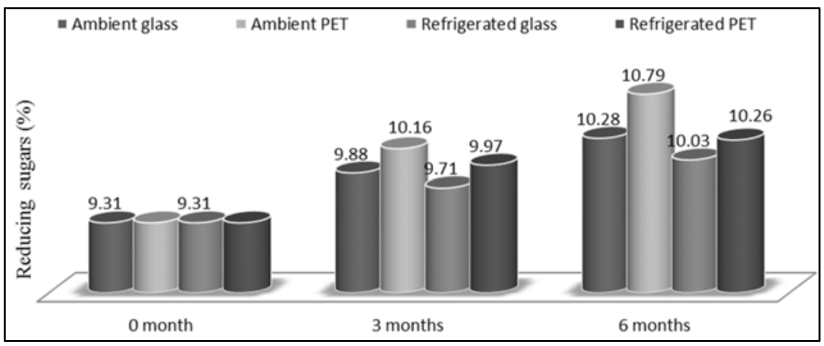

Fig 4: Effect of packaging and storage on reducing sugars (%) of wild aonla drink

Titratable acidity and pH

The titratable acidity of drink showed a slight decrease during storage (Fig. 5) and this decrease might be due to co-polymerization of organic acids with sugars and amino acids. The pH of drink increased slightly during storage which might be due to the degradation of acid in the product during storage. However, the increase was lower in refrigerated storage conditions (increased from 4.20 to 4.25 and 4.27 in both glass and PET) than ambient conditions (increased from 4.20 to 4.28 and 4.30 in both glass and PET). The findings of present studies are in agreement with the results of earlier investigations reported by Thakur *et al.* (2011) [13] in wild pomegranate drink and Hamid *et al.* (2017) [9] in mulberry RTS beverage.

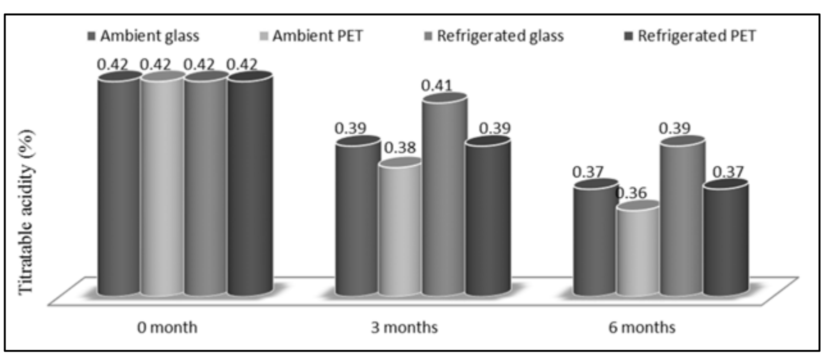

Fig 5: Effect of packaging and storage on titratable acidity (%) of wild aonla drink

Ascorbic acid and total phenols

Decrease in ascorbic acid content during storage (Table 4) might be due to its degradation into dehydro-ascorbic acid or furfural. Ascorbic acid is highly sensitive to heat; therefore its degradation was more in ambient conditions. Significant decrease in total phenol content during storage (Table 5) might be due to their involvement in the formation of polymeric compounds complex formation of phenols with protein and their subsequent precipitations. However, decrease was significantly lower under refrigerated conditions as compared to ambient conditions. Slower rate of loss of phenols and ascorbic acid in refrigerated storage might be due to slower reaction rate in refrigerated conditions as compared to ambient. However, retention of more total phenols and ascorbic acid of drink in glass bottle may also be because of slower reaction rates in glass bottle, as glass material absorb heat at slower rate as compared to PET. Similar findings have been reported by Kumar *et al.* (2010) [15] in aonla drink during storage and Thakur *et al.* (2017) [11] in box-myrtle drink.

Table 4. Effect of packaging on ascorbic acid content (mg/100 g) of wild aonla drink during storage

	V							
	Ambient storage (Months)			**Mean**	**Refrigerated storage (Months)**			**Mean**
S / T	**0**	**3**	**6**		**0**	**3**	**6**	
T_1	61.38	29.84	22.52	37.91	61.38	42.23	38.85	47.49
T_2	61.38	23.08	15.57	33.34	61.38	40.91	35.27	45.85
Mean	61.38	26.46	19.05		61.38	41.57	37.06	
Mean (V)	35.63				46.67			
					$CD_{0.05}$			
T	**SxT Interaction Table**						T= Packaging material	
	0	**3**	**6**	**Mean(T)**	V=0.25	V×S= 0.43	S= Storage period	
T_1	61.38	36.04	30.69	42.70	S= 0.30	V×T= 0.35	V= Storage conditions	
T_2	61.38	31.99	25.42	39.60	T= 0.25	S×T= 0.43	T_1= Glass bottle	
Mean (S)	61.38	34.01	28.05			V×S×T=0.61	T_2= PET bottle	

Table 5: Effect of packaging on total phenols content (mg/g) of wild aonla drink during storage

	V							
	Ambient storage (Months)			**Mean**	**Refrigerated storage (Months)**			**Mean**
S / T	**0**	**3**	**6**		**0**	**3**	**6**	
T_1	2.17	1.86	1.60	1.88	2.17	1.95	1.73	1.95
T_2	2.17	1.73	1.42	1.77	2.17	1.79	1.65	1.87
Mean	2.17	1.80	1.51		2.17	1.87	1.69	
Mean (V)	1.83				1.91			
					$CD_{0.05}$			
T	**SxT Interaction Table**						T= Packaging material	
	0	**3**	**6**	**Mean(T)**	V= 0.03	V×S= 0.06	S= Storage period	
T_1	2.17	1.91	1.67	1.91	S= 0.04	V×T= NS	V= Storage conditions	
T_2	2.17	1.76	1.54	1.82	T= 0.03	S×T= NS	T_1= Glass bottle	
Mean (S)	2.17	1.83	1.60			V×S×T=NS	T_2= PET bottle	

Effect of storage on sensory characteristics of wild aonla drink

The colour, body, taste, aroma and overall acceptability scores of drink decreased significantly during storage (Fig. 6-7) and this decrease was more pronounced under ambient storage conditions than refrigerated storage conditions. Decrease in colour scores during storage might be due to browning caused by copolymerization of organic acids of the product and this might have led the judges to award the lower scores during storage. The possible reason for decrease in body scores might be due to the formation of precipitates in the product as a result of interactions between phenols and protein as well as the formation of cation complexes with phenols during storage. The possible reason for decrease in taste scores might be due to the loss of sugar-acid blend responsible for taste during storage. The decrease in aroma scores during storage might be due to degradation of aromatic compounds in the product. There was a decrease in overall acceptability scores of drink during storage, which might be due to the loss in appearance, flavour compounds and uniformity of the product. Drink packed in glass bottles retained more sensory scores than PET bottles. The retention of better overall sensory scores of drink in glass bottles might be due to the better retention of above given factors as a result of slower reaction rate in glass bottles as compared to PET bottle as a result of their thermal conductance properties. Retention of higher sensory scores in refrigerated conditions might be due to the better condition of the drink during storage as a result of slower rate of chemical reactions. Our results were in conformity with the finding of Thakur *et al.* (2011) [13] in wild pomegranate drink, Hamid *et al.* (2017) [9] in mulberry RTS beverage and Thakur *et al.* (2017) [11] in box-myrtle drink.

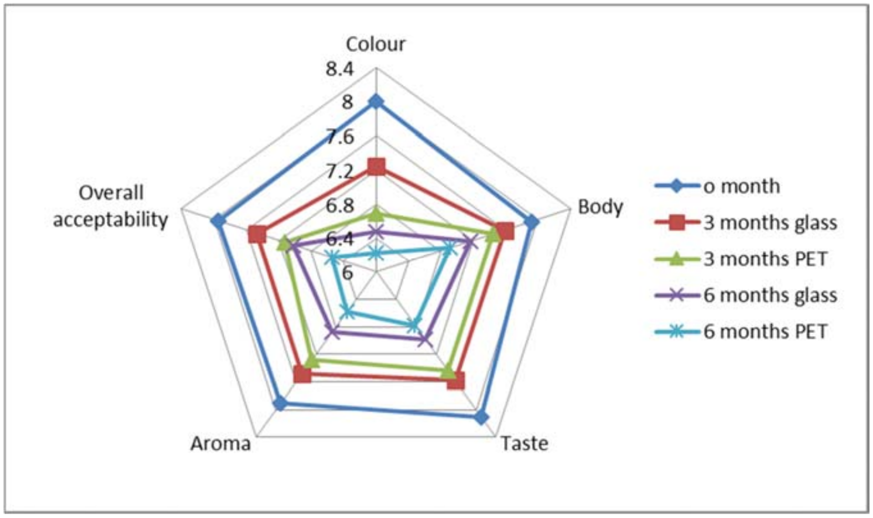

Fig 6: Effect of storage and packaging on sensory characteristics of wild aonla drink stored in glass and PET bottles under ambient storage conditions

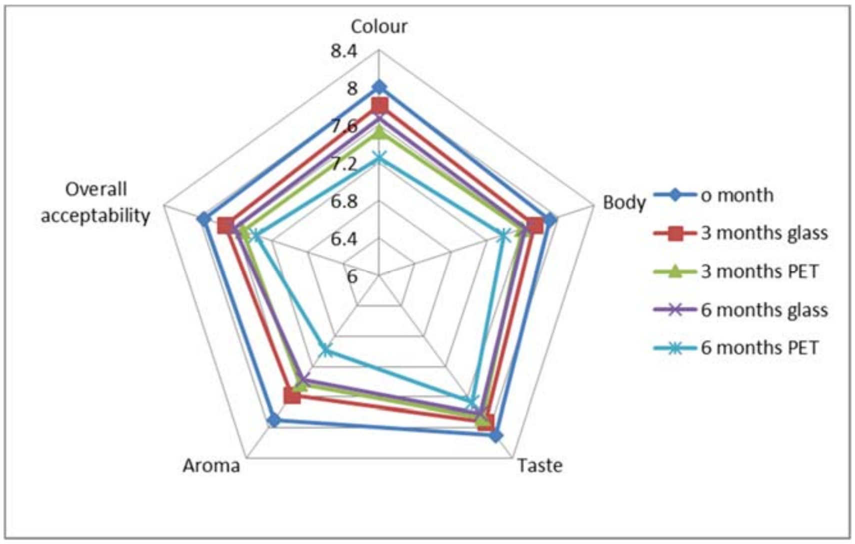

Fig 7: Effect of storage and packaging on sensory characteristics of wild aonla drink stored in glass and PET bottles under refrigerated storage conditions

Conclusion

Eight treatment combinations for the preparation of wild aonla drink were tried and drink with 14 per cent juice, 15 ºB TSS (T_7) and 0.42 per cent acidity was found to be best on the basis of its some physico-chemical and sensory parameters. Drink could be stored safely for a period of six months under both storage conditions and also in both packaging materials with minimum changes in chemical and sensory attributes. However, comparatively minimum changes in drink packed in glass bottle and stored under refrigerated storage conditions were observed as compared to PET bottle. By preparing this type of products this underutilized crop can be exploited in the preparation of a variety of good quality and nutritionally enriched processed products at remunerative cost.

References

1. Parmar C, Kaushal MK. Wild Fruit of Sub-himalayan Region. Kalayani Publisher, New Delhi. 1982, 136.
2. Nath V, Singh IS, Kumar S. Evaluation of aonla cultivars for their shelf-life at ambient temperature. Narendra Deva Journal of Agricultural Research. 1992; 7(1):117.
3. Singh O, Singh R. Development and Evaluation of aonla based blended nectar drink from different fruits using stevia for low calorie. Plant Archives. 2014; 14(1):115-119.
4. Ranganna S. Handbook of analysis and quality control for fruit and vegetable products. Tata McGraw Hill, New Delhi. 2009, 1112.
5. Singleton VL, Rossi JA. Colorimetry of total phenolics with phosphomolybdic phosphotungstic acid reagent. American Journal of Enology and Viticulture. 1965; 16:144-158.
6. Amerine MA, Pangborn RM, Roessler EB. Principles of sensory evaluation of food. Academic Press, London, 1965, 236-268.
7. Cochran WR, Cox CM. Experimental design. John Wiley and Sons, New York. 1967, 171-217.
8. Mahony MO. Sensory evaluation of food: statistical methods and procedures. Marcel Dekker, New York, 1985, 168-169.
9. Hamid, Thakur NS, Kumar P, Thakur A. Studies on preparation and preservation of ready-to-serve (RTS) beverage from underutilized mulberry (*Morus alba* L.)

fruits and its quality evaluation during storage. International Journal of Current Microbiology and Applied Sciences. 2017; 6(9):1067-1079.
10. Bal LM, Ahmad T, Senapati AK, Pandit PS. Evaluation of quality attributes during storage of guava nectar cv. Lalit from different pulp and TSS ratio. Food Processing and Technology. 2014; 5(5):349-353.
11. Thakur A, Thakur NS, Kumar P. Preparation of *Myrica nagi* (Box myrtle) drink and effect of storage temperature on its quality. Journal of Applied and Natural Science. 2017; 9(4):2137-2142.
12. Gould WA. Tomato production, processing and quality evaluation. 2nd ed. Avi Publishing Company, West port, CJ, 1983.
13. Thakur NS, Dhaygude GS, Joshi VK. Development of wild pomegranate drink and its evaluation during storage. International Journal of Food and Fermentation Technology. 2011; 1(2):237-246.
14. Heikal HA, Wakeil EL, Foda IO, Ashmawi H. Preservation of lemon juice. Agriculture Research Review. 1964; 42:68.
15. Kumar S, Godara RK, Sharma RK, Kumar S. Quality evaluation of ready to serve (RTS) from different blends of aonla. Haryana Journal of Horticulture Science. 2010; 39(3, 4):260-262.