The Pharma Innovation Journal 2022; 11(2): 1667-1671

www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277- 7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2022; 11(2): 1671-1671

www.thepharmajournal.com
Received: 06-11-2021
Accepted: 12-12-2021

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Preservation of RTS beverage blended with carrot juice and honey

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Abstract
The study was conducted for the development of refreshing, thirst quenching & energizing ready to serve beverage in which honey can be added in fresh carrot juice that improve the health as well as fulfils the nutritional requirements. The formulation was prepared by combination of different proportions of carrot juice and honey in the ratio of (100:0), (90:10), (80:20) and (70:30) represented by T0, T1, T2 and T3 respectively. The products were subjected to standard physico-chemical, sensory and microbial analysis and accordingly, there was less variation in Overall acceptability scores so all compositions of RTS were acceptable. T0 was found to be the most preferred variant. The storage stability studies showed that the physico-chemical and sensory quality of the RTS blends were acceptable up to 90 days of storage. Microbial analysis of the RTS during storage period up to 90 days revealed that it was free from any spoilage.

Keywords: Honey, carrot juice, ready to serve beverage, overall acceptability, sensory quality, energizing

Introduction
The global juice market has experienced a major growth in recent years, driven by a growing interest of consumers for healthy and nutritionally rich diet (Kahraman and Feng, 2020) [11]. Fruit based beverages like ready to serve (RTS) drink is popular among every age group which are easily digestible, highly refreshing, thirst quenching, appetizing and nutritionally far superior to many synthetic and aerated drinks. Ready to serve (RTS) is a type of fruit beverage which contains at least 10% fruit juice and 10% total soluble solids besides about 0.3% acid. It is not diluted before serving, hence it is known as ready to serve (RTS) (Srivastava and Kumar, 2002: Divyasree *et al.*, 2018) [19, 7].

In recent years, there has been a growing interest in using herbal products as dietary adjuncts in the Food Industry. The development of novel RTS blends is required for meeting the demands of the consumers and also for the growth of food processing industry. Among the new juice products, blended juices have become popular which are prepared by blending of fruits, vegetables and herbal products (Chandra *et al.*, 2018) [5] due to their rich nutrients, unique flavors, and varieties (Kahraman and Feng, 2020) [11].

Ready-to-serve (RTS) beverages made up of fruit pulp have greater amount of water that is useful for body balancing by preventing dehydration. Fruit drinks contain high percentage of sugar and provide a few vitamins and minerals. The consumption of fruit-based beverages in the form of fruit blends is increasing due to public awareness on the presence of various functional ingredients beneficial to health (Sulochanamma *et al.*, 2018) [20].

Carrot is a root vegetable it belongs to *Apiaceae* family and botanical name *Daucus carota* and Genus *Daucus*. Carrot is globally important vegetable crop that provides essential bioactive constituents such as carotenoids, anthocyanins and other phenolic compounds. Carrot supply significant dietary fiber and nutrients in the form of phytochemicals. Carrot also supply significant amount of dietary vitamin A intake through α-and β carotene (Vu Nguyen and Loc Thai Nguyen; Nalinde *et al.*, 2018) [8]. Vitamin A, a fat soluble vitamin, is present abundantly in foods of animal origin such as milk, butter, egg, liver and fish. These foods are expensive and beyond the reach of the poor. However, green leafy vegetables constitute an important item of human diet. Green leafy vegetables and yellow fruits are rich source of beta- carotene, are comparatively less expensive (Devadas *et al.*, 1980) [6]. The carrot roots are the unique roots rich in carotenoids and have a characteristic flavor due to the presence of terpenoids and polyacetylenes (Haq and Prasad, 2014).

Honey is a sweet liquid which is obtained by honey bee *(Apis mellifera)* from nectar of flowering plants which require insect pollination by honey bee. Honey is obtained in approximately every corner of the world and India has been known as 'land of honey' (Puja *et al.,* 2020) [14]. Among natural sweeteners honey is nutritionally a high energy carbohydrate food considered to be the best source of heat and energy, giving over 3,200 calories/kg. About 95% of the honey dry matter is composed of carbohydrates, mainly fructose and glucose. About 5–10% of the total carbohydrates are oligosaccharides, in total about 25 different di- and trisaccharides. Besides, honey contains small amounts of proteins, enzymes, amino acids, minerals, trace elements, vitamins, aroma compounds and polyphenols. Honey has been shown to possess antimicrobial, antiviral, antiparasitory, antiinflammatory, antioxidant, antimutagenic and antitumor effects. Honey can be utilized in the processing of various fruits by replacing the white sugar or jiggery completely or by adding to the product in parts. Its application with various selected fruits that have medicinal properties to produce designer foods (Bogdanov *et al.,* 2003; White, 1975; Ahmad, and Kumaran, 2015) [4, 22, 2].

Materials and Methods

Experiments were conducted to study the development of RTS beverages of carrot juice mixed with honey and its quality evaluation during storage were conducted in the Food Analysis Laboratory and (PG) laboratory in Department of Agricultural Engineering, College of Post-Harvest and Technology, S.V.P. University of Agriculture & Technology, Meerut. For the RTS beverages fresh carrot free from insects and diseases and honey were procured from the local market of Meerut, Uttar Pradesh and brought to the laboratory of S.V.P. Meerut for preparation of ready to serve drinks.

Preparation of ready to serve drink (RTS)

Good quality carrot were procured from the local market and washed into the fresh water so as to remove the dust and dirt particles. Peeling was done manually. The peeled carrot blended in an electrical blender and the juice was filtered through muslin cloth to get clear juice. And then amount of honey is added into juice in the ratio of 100:0, 90:10, 80:20 and 70:30. The requisite amount of sugar and citric acid were dissolved in water to prepare sugar syrup up to boiling stage, and strained through the muslin cloth and added after cooling, to get the desired TSS of 14˚Brix -20˚Brix. The combined carrot juice and honey were then mixed with the sugar syrup in the correct quantity to get the RTS beverage. All the ingredients were dissolved through homogenizing, and heated at 85 °C for 20 minutes. It was removed from the fire and allowed to cool for 10 minutes. Subsequently 70 ppm potassium meta-bi-sulphite (KMS) was added and mixed well with the solution. Prepared RTS beverage was poured into pre-sterilized 200ml capacity glass bottles and capped with stopper immediately, leaving 1" head space on the top and closed with caps air tightly. The sealed bottles were sterilized in hot water bath at 80 °C for 30 minutes. The bottles were removed from the water bath and allowed to cool. The RTS bottles was labeled and stored at refrigerator at 5-10 °C up-to 90 days.

The RTS beverages combinations were

T0 - 100:0 carrot juice and Honey (control) T1 - 90:10 carrot juice and Honey
T2 - 80:20 carrot juice and Honey T3 - 70:30 carrot juice and Honey

Physico-chemical analysis

The prepared RTS beverage formulations blend of Carrot juice with honey were analyzed initially for the parameters of TSS, Titrable acidity, pH, Optical density and Ascorbic acid using AOAC methods. The TSS of each sample was analyzed using hand held refractometer and the values were expressed as °Brix. The acidity in each sample was determined according to standard procedure given in AOAC (2002) [3]. The pH of each sample was recorded with the help of digital pH meter. The ascorbic acid content was estimated by using detective dye, DCPIP (2, 6-dichlorophenol indophenol) according to the standard method of AOAC (2002) [3] and optical density was determined according to standard procedure given in AOAC (2002) [3]. Analyses were carried out for all the formulations (samples) for each parameter and the analyses were replicated thrice.

Sensory evaluation

Sensory quality attributes *viz*. Color, texture, flavor, taste and overall acceptability of the samples were evaluated. Nine points Hedonic rating test method as recommended by Ranganna was used for the purpose of sensory evaluation. This test measures the consumer's acceptability.

Microbial study

Microbial analysis was done to determine the total plate count (TPC) of the samples on Nutrient Agar media and potato dextrose agar (PDA) for yeast and molds counts as method recommended by Harrigan and McCance (1966) [9].

Statistical analysis

Statistically the data was analyzed by using ANOVA with two factors (treatment and storage) and mean were seperated by Ducan's multiple range Test (DMRT) test at 0.05% significant level. The statistical package used for the above analysis was Excel stats.

Result and Discussion

Effect on TSS

Table 1 shows the effect of storage and treatments on TSS of RTS prepared from blend of carrot and honey. According to DMRT, both storage and treatments had significant ($p< 0.05$) effect on TSS (total soluble solids) of ready to serve drinks. From the data this was observed that TSS of treated sample were in range of 12.8°brix to 14.1°brix on zero day of storage, which increase upto14°brix to 15.9°brix at 30 days of storage. The highest mean value (15.1°brix) of TSS in RTS was observed in treatment T3 and lowest in T0 (13.2°brix) and the value of TSS in other treatment are T1 (13.9°brix) and T2 (14.5°brix). It was observed that the value of TSS in RTS was increased with increase in proportions of honey and also increased during storage period of 30 days but after 30 days TSS decreased in all the sample and after 60 days TSS again increased. The total soluble solids tended to increase significantly after one month of storage that might be due to the hydrolysis of polysaccharides into monosaccharide and oligosaccharides. Similar results were also reported by Singh *et al.,* 2014 in ginger and honey blended RTS and ullah *et al.,* 2015 in RTS blended by carrot and kinnow found an

increasing trend in total soluble solids during storage at ambient and low temperature.

Table 1: TSS (°Brix) of carrot juice and honey blend rts during storage

Treatment	Storage Intervals				Means
	0	30	60	90	
T0	12.8±0	14±0	13±0	13.3±0	13.2c
T1	13.2±0	14.5±0	14.2±0	14±0	13.9bc
T2	13.9±0	14.9±0	14.7±0	14.5±0	14.5ab
T3	14.1±0	15.9±0	15.4±0	15±0	15.1a

Figures having different small letters shows significant difference ($p< 0.05$)

Effect on pH

The effect of storage and treatments on pH of value added to ready to serve RTS drink prepared from blend of carrot and honey showed in Table 2. According to DMRT, both storage and treatments had significant ($p<0.05$) effect on pH of ready to serve drinks. This was observed that pH of treated sample were in range of 3.73 to 3.82 on zero day of storage, which decrease upto 3.20 to 3.37 at 90 days of storage. The highest mean value (3.597) of pH in RTS was observed in treatment T0 and lowest in T3 (3.437) and the value of pH in other treatment are T1 (3.547) and T2 (3.497). It is observed that the value of pH in RTS was decreased with increase in proportions of honey and also decreased during storage period of 90 days in all the sample. The degradation of reducing sugar and formation of acidic compounds from it causes a decline in pH. Similar results were also observed by Singh *et al.*, 2014 in ginger and honey blended RTS and Afreen *et al.*, (2016) that pH of all blend of RTS formulation decreased slightly as the storage period proceeded.

Table 2: pH of carrot juice and honey blend RTS during storage

Treatment	Storage Intervals				Means
	0	30	60	90	
T0	3.82±0.030	3.72±0.020	3.50±0.005	3.37±0.058	3.597a
T1	3.79±0.058	3.68±0.01	3.45±0.030	3.22±0.01	3.547ab
T2	3.75±0.015	3.63±0.058	3.39±0.01	3.27±0.026	3.497bc
T3	3.73±0.020	3.52±0.026	3.30±0.025	3.20±0.030	3.437c

Figures having different small letters shows significant difference ($p< 0.05$)

Effect on Acidity

The impact of storage and treatments on acidity of value added to ready to serve RTS drink prepared from blend of carrot and honey showed in Table 3. According to DMRT, both storage and treatments had significant ($p< 0.05$) effect on acidity of ready to serve drinks. This was observed that acidity of treated sample were in range of 0.035 to 0.027 on zero day of storage, which increase upto 0.045 to 0.033 at 90 days of storage. The highest mean value (0.04) of acidity in RTS was observed in treatment T0 and lowest in T3 (0.03) and the value of acidity in RTS in other treatment are T1 (0.0372) and T2 (0.0337). It is observed that the value of acidity in RTS was increased with increase in proportions of honey and also increased during storage period of 90 days in all the sample. The same result were found by Jan and Masih (2012) who concluded increase in acidity during storage which might be effected by the presence of micro-organisms and sugar degradation. It is also an important characteristics which effect on the flavour and overall acceptibility of juice.

Table 3: Acidity of carrot juice and honey blend RTS during storage

Treatment	Storage Intervals				Means
	0	30	60	90	
T0	0.027±0.004	0.028±0.009	0.032±0.004	0.033±0.006	0.04a
T1	0.029±0.006	0.032±0.002	0.035±0.002	0.039±0.004	0.0372ab
T2	0.032±0.006	0.035±0.004	0.040±0.006	0.042±0.002	0.0337c
T3	0.035±0.002	0.038±0.004	0.042±0.009	0.045±0.004	0.03d

Figures having different small letters shows significant difference ($p< 0.05$)

Effect on Optical density

According to DMRT, both storage and treatments on optical density of ready to serve RTS drink prepared from blend of carrot juice and honey had significant ($p< 0.05$) effect showed in Table 4. This was observed that optical density of treated sample were in range of 0.038 to 0.064 on zero day of storage, which increase upto 0.058 to 0.084 at 90 days of storage. The highest mean value (0.0727) of optical density in RTS was observed in treatment T0 and lowest in T3 (0.0467) and the value of optical density in other treatment are T1 (0.0385) and T2 (0.0432). It is noticed that with respect to T1 value of optical density in RTS was increased with increase in proportions of honey and also increased during storage period of 90 days in all the sample sealed in glass bottles during 90 days storage period but in control (T0) their is highest value of optical density 0.064 at freshly prepared RTS and 0.084 at 90 days of storage as compared to honey blended samples due to dark color of carrot and in other composition small amount of honey is used in carrot juice for preparing RTS so the color becomes lighter as compared to control but

O.D is increased as the incorporation of honey increased i.e. T1 (0.030) and T3 (0.038) at freshly prepared RTS and at 90 days the value is increase upto T1 (0.049) to T3 (0.058). The results are similar to the previous study which showed increase in optical density during storage (Yadav *et al.*, 2012).

Table 4: Optical density of carrot juice and honey blend RTS during storage

Treatment	Storage Intervals				Means
	0	30	60	90	
T0	0.064±0.001	0.068±0.0011	0.075±0.0005	0.084±0.0017	0.0727a
T1	0.030±0.0005	0.034±0.002	0.041±0.0017	0.049±0.0015	0.0385d
T2	0.035±0.002	0.038±0.0015	0.045±0.0015	0.055±0.0005	0.0432c
T3	0.038±0.0015	0.042±0.001	0.049±0.002	0.058±0.001	0.0467b

Figures having different small letters shows significant difference ($p< 0.05$)

Effect on Ascorbic acid

Ascorbic acid plays an important role in functional beverage. The changes of Vitamin content in carrot juice blend with honey RTS beverage is shown in Table 5. The ascorbic acid content of the ready to serve drink prepared from blend of carrot juice and honey decreased during storage due to the fact that ascorbic acid being sensitive to oxygen, light and heat was easily oxidized in presence of oxygen by both enzymatic and non-enzymatic catalyst (Mapson, 1970). The ascorbic acid also decreased significantly ($p< 0.05$) from 12.2 to 8.6 mg/100 ml with an increase in the concentration of honey from 10 to 30% in the RTS beverage formulations. According to DMRT, both storage and treatments had significant ($p< 0.05$) effect on ascorbic acid of ready to serve drinks. This was observed that ascorbic acid of treated sample were in range of 12.2 to 8.6 on zero day of storage, which decrease upto 5.0 to 3.0 at 90 days of storage. The highest

mean value (8.8) of ascorbic acid was observed in treatment T0 and lowest in T3 (0.03) and the value of ascorbic acid in other treatment are T1 (0.0372) and T2 (0.0337). Maximum ascorbic acid content was found in RTS T0 at day 0 which gradually decreased till day 90. Minimum ascorbic acid content was found in RTS T3 at the day 90.

Table 5: Ascorbic acid of Carrot Juice and Honey Blend RTS during Storage.

Treatment	Storage Intervals				Means
	0	30	60	90	
T0	12.2±0.2	10.1±0.11	7.9±0.20	5.0±0.2	8.8a
T1	11.5±0.11	9.3±0.1	6.8±0.11	4.3±0.11	7.97ab
T2	9.9±0.1	7.0±0.20	5.4±0.1	3.7±0.11	6.5bc
T3	8.6±0.20	6.6±0.2	4.2±0.11	3.0±0.1	5.6c

Figures having different small letters shows significant difference ($p< 0.05$)

Sensory evaluation

Overall sensory scores obtained by the different beverages are documented in Table 6 which shows the evaluated results of the different sensory properties, namely, color, taste, flavor, mouth feel and overall acceptability for the juices. The overall acceptability was taken as the average of the four parameters (color, taste, flavor, mouth feel and overall acceptability). The sensory data for change in overall acceptability score of carrot and honey RTS during storage sealed in glass bottles are presented in Table 6. The score for overall acceptability ranged from 7.84 – 7.99. The highest value was observed for C100H0 (7.99) and the lowest for C70H30 RTS (7.84). Highest overall acceptability score was evaluated for C100H0 RTS (7.99) followed by C90H10 (7.94), C80H20 (7.90) and C70H30 (7.84). There was less variation in O.A scores so all compositions of RTS were also acceptable. From the perusal of data that overall acceptability scores was decreased with increase storage period among all the samples sealed in glass bottles during storage. The overall acceptability score decreased for C100H0 (7.99 to 7.82), C90H10 (7.94 to 7.77), C80H20 (7.90 to 7.73) and C70H30 RTS (7.84 to 7.70) stored under refrigeration condition from fresh to 90 days of storage, respectively. There was noticeable decrement in overall acceptability with increasing the incorporation of honey.

Table 6: Sensory acceptance of carrot juice and honey blend RTS during storage

Treatment	Storage Intervals				Means
	0	30	60	90	
T0	7.99±0.55	7.93±0.58	7.87±0.56	7.82±0.56	7.902a
T1	7.94±0.56	7.89±0.54	7.82±0.54	7.77±0.57	7.855b
T2	7.90±0.57	7.86±0.57	7.78±0.56	7.73±0.58	7.817c
T3	7.84±0.59	7.80±0.59	7.74±0.59	7.70±0.59	7.77d

Figures having different small letters shows significant difference ($p< 0.05$)

Microbiological analysis

The RTS beverage was observed to be microbiologically safe as the growth of bacteria, yeast and molds were found negligible on the day of preparation and no further growth during storage for a period of three months. The microbial analysis of the carrot based RTS formulations were performed at 30, 60 and 90 days after storage. Treatments from any microbial growth till the end of storage period in terms of bacterial plate count indicates that the RTS formulations can be stored without any microbial damage till 90 days after preparation, thereby indicating its fitness for consumption even after 3 months of storage. The absence of the microbial count could be because of pasteurization and particular use of citric acid and KMS as preservative. The results was observed similar with the findings of (Ramachandran and Nagarajan 2014: Sulochanamma *et al.*, 2018) [18, 20].

Conclusion

The RTS beverages prepared by using carrot juice and honey was nutritious and also safe for consumption since there is no addition of artificial color and flavor. RTS beverage was free from any microbial contamination. The RTS beverage C100H0 ratio was observed highest value of pH and optical density and C70H30 sample was observed highest value of TSS, acidity as fresh and during storage of 30, 60 and 90 days and optical density was also highest in C70H30 as compared to C90H10. The study also showed that the value of acidity was increased and TSS was increased at 30 days and after 30 days the value of TSS was decreased with increasing storage period and there was noticeable decrement in overall acceptability with increasing the incorporation of honey with carrot juice RTS beverage was packed in glass jar and stored at refrigeration condition (5 °C).

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