

Studies on development and storage of whey-based pineapple (*Ananas comosus*) and bottle gourd (*Lagenaria siceraria*) mixed herbal beverage

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Abstract

Whey-based pineapple and bottle gourd mixed herbal (WPBH) beverage was prepared with the incorporation of *Mentha arvensis* extract (0 to 4%). The storability of the beverage was studied at 7±10C for 20 days. 10 ml each of pineapple and bottle gourd juice and 8 g of sugar were fixed per 100 ml of the herbal beverage. Whey quantity varied from 68 to 72 ml for 100 ml of the beverage depending upon the concentration of *Mentha* extract. The organoleptic scores and overall acceptability of the beverage improved with increase in *Mentha* extract from 0 to 2%. Addition of 3 and 4% *Mentha* extract decreased the beverage quality as beverage scored lower organoleptic scores. Acidity and TSS (Total Soluble Solids) content increased while pH decreased during storage. A significant decrease in ascorbic acid content was also observed during storage. The overall acceptability of the beverage was desirable up to 15 days of storage at refrigeration temperature. Whey based beverage prepared from pineapple and bottle gourd juices in combination with edible extract of herbal medicinal plants like *Mentha arvensis* will not have only excellent nutritional properties but will also possess therapeutic, prophylactic, antibacterial and organoleptic properties.

Keywords

Herbal beverage
whey
pineapple
bottle gourd
sensory quality
storage

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Introduction

Whey is a nutritious by product from cheese, chhana and paneer industry containing valuable nutrients like lactose, proteins, minerals and vitamins etc. which have indispensable value as human food. Whey constitutes 45-50% of total milk solids, 70% of milk sugar (lactose), 20% of milk proteins and 70-90% of milk minerals and most importantly, almost all the water soluble vitamins originally present in milk (Horton, 1995). In India, it is estimated that about 100 million kg of whey is annually derived as a byproduct which may cause substantial loss of about 70,000 tonnes of nutritious whey solids (Parekh, 2006). Considerable work has been done throughout the world to utilize whey for production of whey protein concentrate (WPC), whey powder, lactose, lactic acid, whey paste etc. (Panesar *et al.*, 2007).

The conversion of whey into beverages through fermentation or without fermentation is one of the most attractive avenues for the utilization of whey for human consumption. In terms of functionality, whey protein enhances protein content of beverage while

improving its quality. The production of a beverage from whey butter cheese and acerola juice has been shown to have good commercialization potential, uniting the benefits provided by the former with those of latter, including the ingestion of essential amino acids and increasing the vitamin C content, resulting in a product of differentiated nutritive value (Cruz *et al.*, 2009). Singh *et al.* (1999) attempted to develop a soft beverage from paneer whey and guava. Sikder *et al.* (2001) formulated different blends of whey beverages by using various levels of mango pulp (8-12%) with 0.04% acidity. Whey-based mango herbal beverage prepared with 2 % *Mentha* extract has been found to exhibit highest overall acceptability on the day of preparation as well as after 30 days of storage (Sirohi *et al.*, 2005). Selected characteristics of whey-fortified banana beverages stored at 4, 20, 30 and 40°C were monitored at specific time intervals over a 60-day storage period. The sensory characteristics of the whey-banana beverage stored at 4°C were studied and the product was a sour, sweet, smooth beverage, with distinctive banana flavor and minimum off-flavor (Ernest *et al.*, 2005). Yadav *et al.*

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(2010) developed whey based banana herbal beverage incorporating mentha arvensis extract varying from 0 to 4 % and reported that the organoleptic score and overall acceptability of the herbal beverage improved with increase in mentha extract concentration up to 2%.

Next to bananas, pineapple is second most popular tropical fruit. It is cheaply available in market. It contains micro-nutrients and the experts believe that it protects against cancer and this micro-nutrient break up blood clots and is beneficial to the heart. The bottle gourd (*Lagenaria siceraria*) is greenish in color, bottle shaped or round shaped vegetable. Bottle guard (*Lagenaria siceraria*) is 96.1% water, so is light on the stomach and aids digestion. It is beneficial for health in many ways. The pulp of the fruit is considered cool, diuretic, antibilious, and useful in coughs and as antidote to certain poisons (Duke, 1992; Ghule et al., 2006; Ghule et al., 2007). Decoction of leaves mixed with sugar is considered beneficial in jaundice and fruit is also used in cholera (Rahman et al., 2008). However, due to poor sensory acceptability, incorporation of bottle guard with other fruits to develop health promoting products is very scanty. Herbal extract of *Mentha arvensis* has preventive and curative value. It is used to treat sour throat, gastric problems and other problems related to gastrointestinal tract (Campbell et al., 1973; Jamal et al., 2006). Whey based beverage prepared from pineapple & bottle gourd juices in combination with edible herbal medicinal plant extract of *Mentha arvensis* will not have only excellent nutritional and sensory properties but will also possess therapeutic, prophylactic, antibacterial and organoleptic properties. Therefore, keeping in view of the nutritional and functional attributes of bottle gourd and pineapple, potential of whey to be used in nutritious and health promoting beverages and efficacy of herbs like mentha arvensis as flavorants and natural preservatives, the present study was undertaken with an objective to develop a value added whey based herbal beverage along with its storage study.

Materials and Methods

This study was carried out in year 2010. The raw materials such as double toned milk (vita brand), pineapple, bottle gourd, *Mentha arvensis* (green leaves) and sugar were purchased from a local market.

Preparation of pineapple juice

Ripe pineapple were selected, the crown and stem portion were removed and the fruit was washed in tap water. They were sliced using mechanical slicer. The

rind was peeled with knives and eyes were removed. The prepared slices were crushed in a mixer grinder and the juice was recovered by pressing the crushed mass in a hydraulic press. The juice was filtered with muslin cloth. The juice was kept at a refrigeration temperature ($7\pm 1^{\circ}\text{C}$) until used.

Preparation of bottle gourd juice

The healthy fruits of uniform size and color were peeled off and cut into small pieces. The juice was extracted in a juicer and filtered through two layers of muslin cloth (Deore et al., 2008).

Preparation of mentha extract

The leaves of *Mentha arvensis* were washed, ground in a mixer grinder and filtered using muslin cloth.

Preparation of whey

The heated milk (84°C) was acidified by adding citric acid (2%) followed by continuous stirring resulting in complete coagulation of milk protein (casein). The liquid whey was filtered using muslin cloth.

Preparation of whey-based pineapple and bottle gourd mixed herbal (WPBH) beverage

WPBH was prepared with the addition of mentha extract varying from 0 to 4% concentration. For the preparation of 100 ml of herbal beverage, whey amount varying from 68 to 72 ml was added with 10 ml of pineapple juice and 10 ml of bottle gourd juice (Table 1). The whey, juices and sugar (8 g/100 ml of beverage) were mixed in the given amount, preheated to 45°C before mixing mentha extract. The beverages obtained were filtered and filled into presterilized glass bottles (200 ml) and sealed. Pasteurization of filled bottles was done in boiling water for 30 min (Lal et al., 1998) and cooled to room temperature.

Storage studies

Bottles containing beverages were stored at refrigerated temperature ($7\pm 1^{\circ}\text{C}$) for 20 days. Samples were drawn at intervals of 5 days and evaluated for physico-chemical and organoleptic properties.

Physico-chemical analysis

The juice samples (pineapple and bottlegourd), whey and herbal beverage samples were analyzed for their different physico-chemical properties. Total soluble solids were determined with refractometer (0-320) and the values were expressed as $^{\circ}\text{Brix}$. Total acidity was calculated in terms of lactic acid for whey and citric acid for juice samples and herbal beverage samples by titrating against 0.1N NaOH according to

AOAC (1995) method. The total sugar content was determined by the method as described by Ranganna (1986). Protein content was determined by Kjeldahl method for nitrogen estimation, using factor of 6.38 for conversion of nitrogen into protein (BIS, 1961). Fat content was determined by Gerber centrifuge method (BIS, 1977). Ascorbic acid was determined by the method as described by Ranganna (1986).

Sensory analysis of WPBH beverage

The sensory analysis was performed on a 9-point hedonic scale by a panel of 20 semi trained members. The beverage samples were evaluated for color, appearance, flavor, taste and overall acceptability. The evaluation was done at an interval of 5 days during the storage period of 20 days.

Statistical analysis

Data obtained on different characteristics were subjected to statistical analysis to find out the effect of different levels of mentha extract and storage period on physico-chemical and sensory characteristics of the product. The data were analyzed statistically in a completely randomized design (CRD) using one factor and two factor analysis of variance (ANOVA) with the help of OPSTAT.

Results and Discussion

Physico-chemical characteristics of whey, pineapple and bottle gourd juice

The physico-chemical characteristics of whey, pineapple and bottle gourd juice are given in Table 2. Whey was used in the preparation of beverage in place of water. The TSS content of whey was higher (5.63%) as compared to pineapple (3.86%) and bottle gourd juice (3.46%). The ascorbic acid content of pineapple was slightly higher as compared to bottle gourd. The total acidity measured as % lactic acid in whey and % citric acid in juice samples was highest in pine apple juice (0.66%) as compared to bottle gourd juice and whey.

Effect of mentha extract concentration and storage period on physico-chemical characteristics of WPBH beverage

The TSS content of freshly prepared WPBH beverage samples of H0 (control), H1, H2, H3 and H4 was measured as 13.93, 14.03, 13.97, 13.77 and 13.730Brix respectively (Table 3). It was observed that the concentration of mentha extract did not affect the TSS content of WPBH appreciably and the values of TSS for different concentration levels of mentha extract were comparable, although this effect was statistically significant ($p < 0.05$). Table 3 shows that

Table 1. Formulations for preparation of 100 ml of WPBH beverage

Treatment	Pineapple juice (ml)	Bottle gourd juice (ml)	Sugar (g)	Mentha extract (ml)	Whey
H ₀	10	10	8	0	72
H ₁	10	10	8	1	71
H ₂	10	10	8	2	70
H ₃	10	10	8	3	69
H ₄	10	10	8	4	68

H0 is control beverage

Table 2. Physico-chemical characteristics of whey, pineapple and bottle gourd juice

Characteristics	Whey	Pineapple	Bottle gourd
TSS ^o Brix	5.63 ± 0.01	3.86 ± 0.03	3.46 ± 0.03
Ascorbic acid (mg/100g of sample)	*	15.66 ± 0.6	14.33 ± 0.3
pH	5.00 ± 0.01	4.79 ± 0.01	5.74 ± 0.03
Acidity (% LA or %CA)	0.39 ± 0.01	0.66 ± 0.06	0.36 ± 0.06
Fat (%)	0.01 ± 0.01	*	*
Protein (%)	0.18 ± 0.02	*	*

CA: Citric acid, LA: Lactic acid

* Not determined

The values are mean of three independent determinations

the storage period had statistically significant effect on the average TSS of the beverages ($p < 0.05$) up to 15 days of storage. The interaction effect between mentha extract concentration and storage period was also significant ($p < 0.10$). The highest value of TSS was recorded to be 14.37 ^oBrix at the end of 15 or 20 days of storage for the beverage containing 1% mentha extract. Increase in TSS might be due to the solubilization of insoluble portion of the products due to presence of acids (ascorbic and citric acid) during storage as reported by Sethi (1992). Barwal *et al.* (2005) also observed an increase in the TSS of the developed bitter gourd RTS drink during storage. Increase in TSS was also reported by Yadav *et al.* (2010) in whey based banana herbal beverage.

Acidity of WPBH beverage samples of H0 (control), H1, H2, H3 and H4 was 0.37, 0.38, 0.37, 0.40 and 0.43% respectively (Table 3). The effect of storage period on the acidity of beverages was found to be significant and the mean value of 0.39% for acidity in case of freshly prepared beverages increased to 0.53% in the samples stored for 20 days. A slight but statistically significant ($p < 0.05$) change in the values of acidity was observed when *Mentha* extract concentration was increased from 0 to 4%. The interaction effect of *Mentha* extract concentration and storage period on acidity was significant ($p < 0.05$). The increase in acidity with storage period was recorded in all the treatments. The increase in acidity was due to conversion of lactose to lactic acid, and formation of organic acid by ascorbic acid present in juice. This increase might also have been attributed to polyphenols present in *Mentha* extract and their degradation. The conversion of proteins into amino

Table 3. Effect of treatment (*Mentha arvensis* concentration) and storage period on TSS (°Brix), acidity (%) and pH of WPBH beverage

Storage period (days)	TSS (°Brix)						Acidity (%)						pH					
	H ₀	H ₁	H ₂	H ₃	H ₄	Mean for storage period	H ₀	H ₁	H ₂	H ₃	H ₄	Mean for storage period	H ₀	H ₁	H ₂	H ₃	H ₄	Mean for storage period
0	13.93	14.03	13.97	13.77	13.73	13.89	0.37	0.38	0.37	0.40	0.44	0.39	4.99	4.96	5.01	5.02	5.01	4.90
5	14.07	14.13	14.27	13.87	13.83	14.03	0.44	0.44	0.43	0.51	0.44	0.45	4.92	4.90	4.99	4.98	4.95	4.97
10	14.17	14.27	14.27	14.07	14.03	14.16	0.46	0.43	0.45	0.46	0.45	0.45	4.84	4.81	4.90	4.91	4.89	4.92
15	14.23	14.37	14.33	14.23	14.17	14.27	0.44	0.44	0.46	0.52	0.48	0.47	4.79	4.73	4.81	4.81	4.85	4.90
20	14.27	14.37	14.26	14.03	14.26	14.24	0.54	0.53	0.57	0.49	0.53	0.53	4.73	4.68	4.75	4.78	4.78	4.80
Mean for treatment	14.13	14.23	14.22	13.99	14.01		0.45	0.44	0.46	0.48	0.47		4.90	4.87	4.94	4.94	4.97	

C.D. for treatment = 0.05

C.D for storage period= 0.05

Interaction C.D. =0.10

The values are the means compared at a significance level of 5%

C.D. for treatment= 0.01

C.D. for storage period= 0.01

Interaction C.D. = 0.02

C.D. for treatment= 0.02

C.D. for storage period= 0.02

Interaction C.D.=0.03

Table 4. Effect of treatment (*Mentha arvensis* concentration) and storage period on total sugars and ascorbic content of WPBH beverage

Storage period (days)	Total sugars (mg/100g of sample)						Ascorbic acid (mg/100g of sample)					
	H ₀	H ₁	H ₂	H ₃	H ₄	Mean for storage period	H ₀	H ₁	H ₂	H ₃	H ₄	Mean for storage period
0	13.12	13.44	13.35	13.48	13.15	13.31	1.36	1.39	1.42	1.45	1.51	1.43
5	13.17	13.42	13.31	13.47	13.16	13.30	1.33	1.35	1.38	1.38	1.42	1.37
10	13.17	13.44	13.45	13.47	13.17	13.34	1.33	1.28	1.33	1.34	1.33	1.32
15	13.17	13.44	13.35	13.49	13.15	13.32	1.30	1.24	1.28	1.29	1.29	1.28
20	13.15	13.44	13.32	13.40	13.17	13.31	1.20	1.19	1.19	1.19	1.24	1.20
Mean for treatment	13.16	13.43	13.36	13.47	13.16		1.30	1.29	1.31	1.33	1.36	

C.D for treatment =0.02

C.D. for storage period=0.02

Interaction C.D. =0.04

Interaction C.D. =0.02

The values are the means compared at a significance level of 5%

C.D. for treatment=0.01

C.D for storage period=0.01

increase in acidity and decrease in pH of beverages. Similar results have also been reported by Kalra *et al.* (1991) and Sikder *et al.* (2001) for mango RTS.

The total sugar content of freshly prepared WPBH beverage samples of H₀, H₁, H₂, H₃ and H₄ were 13.12, 13.44, 13.35, 13.48 and 13.15 respectively (Table 4). The storage period had no significant effect on the total sugar content of beverages and the mean value of 13.31% for freshly prepared beverages remained almost same after 20 days of storage ($p < 0.05$). The results are in agreement with those of Sirohi *et al.* (2005) who also observed no variation in the total sugars content during the storage of whey-based mango herbal beverage. Yadav *et al.* (2010) also observed no variation in total sugar content during the storage of whey-based banana herbal beverage. However, the results of the present study are in contradiction with those of Barwal *et al.* (2005) and Kumar and Manimegalai (2005) who observed a decrease in the total sugars content during storage of RTS from bitter gourd and whey-based papaya RTS respectively. However, the storage period in these studies was much higher i.e. 180 and 90 days respectively which could have favored the Maillard reaction and other chemical reaction of sugars with acids during the storage resulting in decrease in total sugar content. The effect of *Mentha* extract concentration was significant on the total sugar content of the beverages ($p < 0.05$). The total sugar content of 13.12% in the control sample (H₀) increased to 13.48%, when *Mentha* extract concentration was increased to 3% (H₃).

The ascorbic acid content of freshly prepared WPBH beverage samples of H₀ (control), H₁, H₂, H₃, and H₄ were 1.36, 1.39, 1.42, 1.45 and 1.51 mg respectively. The storage period had a highly significant ($p < 0.05$) effect on the ascorbic acid content of beverages and the mean value was 1.43 for freshly prepared beverage and decreased to 1.20 after the storage of 20 days as shown in Table 4.

acids during storage is also possible. The results are in agreement with the findings reported by Soliman *et al.* (1995), Sikder *et al.* (2001) and Yadav *et al.* (2010).

The pH value for freshly prepared WPBH beverage samples of H₀ (control), H₁, H₂, H₃ and H₄ were 4.99, 4.96, 5.01, 5.02 and 5.01 respectively as shown in Table 3. Storage period had a significant decreasing effect on the pH of the beverages with a mean value of 4.90 for freshly prepared beverages and 4.80 for beverages stored for 20 days ($p < 0.05$). The *Mentha* extract concentration also affected the pH of the beverages significantly ($p < 0.05$). The interaction effect of *Mentha* extract concentration and storage period on pH was also significant ($p < 0.05$). With the increase in storage period, the pH of all treatments decreased. This fact is in accordance with the results obtained for acidities of different beverage samples as with increase in acidity pH correspondingly decreases. This may be due to the production of organic acids and amino acids due to the action of ascorbic acid on sugar and protein content of the beverages. Lactose and proteins are converted into lactic acid and amino acids leading to

Table 5. Effect of treatment (*Mentha arvensis* concentration) and storage period on color and appearance of WPBH beverage

Storage period (days)	Color						Appearance					
	H ₀	H ₁	H ₂	H ₃	H ₄	Mean for storage period	H ₀	H ₁	H ₂	H ₃	H ₄	Mean for storage period
0	8.00	8.00	8.50	8.00	8.00	8.10	8.00	9.00	9.00	9.00	8.00	8.60
5	7.00	8.00	8.50	8.00	8.00	7.90	8.00	8.00	8.00	8.00	7.50	7.90
10	7.00	8.00	7.50	8.00	7.00	7.50	7.50	7.00	8.00	7.00	7.50	7.40
15	7.00	7.00	7.50	7.00	7.00	7.10	7.00	6.00	7.00	6.00	7.00	6.60
20	6.00	6.00	6.50	7.00	6.50	6.40	6.00	6.00	7.00	5.00	5.50	5.90
Mean for treatment	7.00	7.40	7.70	7.60	7.30		7.30	7.20	7.80	7.00	7.10	

C.D. for treatment= 0.31

C.D. for treatment= 0.26

C.D. for storage period= 0.31

C.D. for storage period=0.26

Interaction C.D. = NS

Interaction C.D.= 0.58

The values are the means compared at a significance level of 5%

The *Mentha* extract concentration also affected the ascorbic acid content of the beverages significantly ($p < 0.05$). Similar result for ascorbic acid was given by Ramasastry (1974), the ascorbic acid decreased from 8.43 to 6.65 mg/100 gm during storage of anola powder. Krishnaveni *et al.* (2001) also reported a decrease in ascorbic acid content with the increase in storage intervals in storage stability of jack fruit RTS beverage.

Effect of mentha extract concentration and storage period on sensory characteristics of WPBH beverage

The storage period significantly decreased the color ratings of the product ($p < 0.05$) and the mean value of 8.10 for color for the freshly prepared beverage decreased to 6.40 after 20 days of storage (Table 5). The *Mentha* extract concentration had a significant effect on color at 4% level ($p < 0.05$) with 2% level of *Mentha* extract concentration showing maximum liking on average basis for color in WPBH. The appearance of the beverages was

affected significantly with storage and it decreased to a value of 5.90 after 20 days from initial value of 8.60 (Table 5). The interaction effect of storage period and *Mentha* extract concentration was non-significant on the appearance. However, the color of H2 WPBH beverage (2% *mentha* extract) was most liked during the storage period. The taste and flavor decreased significantly with storage period and the mean value of 7.50 for flavor and taste in the freshly prepared beverage decreased respectively to 5.90 and 4.90 after 20 days of storage (Table 6). The decrease in the flavor during storage could be possibly due to loss of volatile aromatic substances (Thakur and Barwal, 1998). The effect of *Mentha* extract concentration on the flavor and taste was significant and at 4% level of concentration, it decreased the flavor and taste significantly ($p < 0.05$). However, 1% (H1) and 3% level of mentha extract respectively resulted in best average score for taste and flavor.

The overall acceptability of the WPBH beverage during 20 days of storage period decreased significantly from 8.02 to 5.80 and the product was slightly desirable up to 15 days having a value of 6.45 for overall acceptability (Table 6). The increase in *Mentha* extract concentration increased overall acceptability significantly up to 1-2% level and after further increase in its concentration, overall acceptability decreased.

Conclusion

From the results of the present study, it can be concluded that a whey based pineapple and bottle gourd herbal (WPBH) beverage can be prepared successfully with the incorporation of 2% *Mentha* extract. The beverage can be stored at refrigeration temperature without adding any chemical preservative

Table 6. Effect of treatment (*Mentha arvensis* concentration) and storage period on flavor, taste and overall acceptability of WPBH beverage

Storage period (days)	Flavor						Taste						Overall acceptability					
	H ₀	H ₁	H ₂	H ₃	H ₄	Mean for storage period	H ₀	H ₁	H ₂	H ₃	H ₄	Mean for storage period	H ₀	H ₁	H ₂	H ₃	H ₄	Mean for storage period
0	7.50	8.00	8.00	8.00	7.00	7.70	8.00	8.00	8.00	7.50	7.00	7.00	7.87	8.25	8.37	8.00	7.62	8.02
5	7.00	7.50	8.00	8.00	6.50	7.40	8.00	8.00	7.00	6.50	6.50	7.20	7.50	7.87	7.87	7.62	7.12	7.60
10	7.00	7.50	7.00	7.50	6.00	7.00	7.00	8.00	7.00	6.00	5.50	6.70	7.12	7.62	7.37	7.12	6.50	7.15
15	6.00	7.00	6.00	7.00	6.00	6.40	6.00	7.00	6.00	5.50	4.50	5.80	6.50	6.75	6.62	6.37	6.00	6.45
20	6.00	6.50	5.00	6.50	5.50	5.90	5.00	6.00	5.00	4.50	4.00	4.90	5.75	6.12	5.87	5.75	5.50	5.80
Mean for treatment	6.70	7.30	6.80	7.40	6.20		6.80	7.40	6.60	6.00	5.50		6.95	7.32	7.22	6.97	6.55	

C.D. for treatment = 0.36

C.D. for treatment= 0.34

C.D. for treatment= 0.15

C.D. for storage period= 0.36

C.D. for storage period= 0.34

C.D. for storage period= 0.15

Interaction C.D. = NS

Interaction C.D. = NS

Interaction C.D.=NS

The values are the means compared at a significance level of 5%

with desirable consumer acceptability up to 15 days. The product can prove a nutritionally as well as organoleptically desirable beverage with agreeable taste, energy providing due to whey proteins in it. Product developers seeking out functional and nutritional attributes of whey to tap the tremendous growth opportunities in the beverage industry can move forward for the development of such herbal beverages based upon whey and other fruits. Pine apple and bottle gourd known for their nutritional and medicinal properties as well recognized by Indian Ayurveda also can be exploited to develop nutritious health drinks with inherent therapeutic properties.

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