



RESEARCH ARTICLE

Development and Evaluation of Whey-based Herbal Beverages as Health Drink

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ABSTRACT

The present study aimed to develop whey-based herbal beverages and to evaluate their physical, chemical, and sensory properties. The pH, titrable acidity (% lactic acid) and total soluble solids (^oBrix) of whey used for the development of whey based herbal drinks were found to be 5.21, 0.23 and 6.4 respectively. The proximate composition of whey such as lactose (g/100g), protein (g/100g) and fat (g/100g) were 4.28, 0.28 and 0.13, respectively. The whey (65%), sugar (11%), H₁ - brahmi extract (3%), H₂ - mint extract (2%) and H₃ - jaljeera powder (0.15%) were standardized for the formulation of whey-based herbal drinks. Among the three variations, whey-based jaljeera drink (H₃) scored the highest sensory acceptability. The pH, titrable acidity (% citric acid), total soluble solids (^oBrix), lactose (g/100g), protein (g/100g), fat (g/100g), total reducing sugar (g/100g), total sugar (g/100g), calcium (mg/100g), ascorbic acid (mg/100g) and total phenol (mgGAE/100g) of whey-jaljeera drink (H₃) were 5.24, 0.21, 12.0, 2.78, 0.46, 0.30, 3.43, 14.43, 55.27, 0.12 and 14.26, respectively. The sensory attributes retention was high in glass (P₂) compared to PET (P₁). The sensory attributes were gradually decreased during the storage of room temperature (R₁) than refrigerated temperature (R₂). The cost of the formulated whey brahmi drink, whey mint drink, and whey jaljeera drinks were Rs.18.33/250ml, Rs.20.20/250ml and Rs.14.75/250ml, respectively. The study concluded that nutritionally rich whey-based herbal drinks can be recommended to all group of people.

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INTRODUCTION

The beverages provide hydration to the body and nourish our body with essential nutrients such as carbohydrates, protein, fat, vitamins and minerals (Abdellatif, 2018). The Indian Natural Healthy Beverages recorded a 22% growth rate in 2016 (MOFPI, 2017). Whey has high protein digestibility corrected amino acid source (PDCAAS), amino Acid (AA), protein efficiency ratio (PER), biological value (BV) and protein digestibility (PD) score 1.00, 1.14, 3.2, 100 and 99% respectively (Gangurde *et al.*, 2011). Whey protein is a versatile food ingredient and had high demand in the novel product development sectors. It has functional properties such as emulsification, texture modifier, carrier/vehicle, surface-active component, foaming, gelation, and water-binding (de Castro *et al.*, 2017). The large volume of whey is produced as a by-product from the manufacturing of cheese/paneer/channa/chakka in dairy industries. It has high BOD (>35,000 ppm) and COD (>60,000 ppm), so it is considered

as the most pollutant by-product from the food sectors. The main component of liquid whey was lactose (Macwan *et al.*, 2016). Solak and Akin (2012) reported that the group of whey proteins consisted of β -lacto globulin, α -lactalbumin, bovine serum albumin, immunoglobulins and others. Gupta and Prakash (2017) reported that carbohydrates in whey had the ability to act prebiotic. IgG is binding the bacterial toxins and reducing the bacterial load in the bowel. The sphingomyelin had the ability to prevent the colon cancer. The glutathione plays critical role in the prevention of free radical damages, pollutant, infecting agent, ultraviolet rays and toxins. The liquid whey is the good raw material for the development of low cost beverages. Whey protein beverages have high popularity, excellent nutritional qualities and ease of digestibility. The whey beverages can be classified into (i) whey incorporated into fruit/vegetable juices (ii) fermented or unfermented thick beverages (iii) thirst quenching beverages and (iv) alcoholic beverages (Chavan *et al.*, 2015b). Brahmi

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(*Bacopa monnieri*) belongs to the Scrophulariaceae family. It had the dietary antioxidant to reduce the free radical formation and help to improve brain functions in humans (Jeyasri et al., 2020). Mint (*Mentha spicata*) belongs to the family of Lamiaceae. It had several biological activities and was used in folkloric medicine as antibacterial, antifungal, antioxidant, antispasmodic, diuretic and for the treatment of colds, flu, respiratory tract problems and stomach-ache (de Sousa Barros et al., 2015). Black cumin (Jaljeera) (*Cuminum cyminum* L.) contains carbohydrate, protein, lipids, minerals and polyunsaturated fatty acids and is used for medical purposes. It had a strong aromatic flavor extensively used in the flavoring industry. It is considered as digestive stimulants, diuretic, asthmatic, galactagogue and emmenagogue. (Mamun and Absar 2018). Perasiriyan et al. (2013) concluded that 1.5% of tea incorporated with whey as a healthy drink had scored highest sensory acceptance upto 3 days at the refrigerated temperature ($7\pm 10^{\circ}\text{C}$). The formulated whey based tea enriched with polyphenols. Maya and Ritu (2016) formulated that whey blended with orange juice beverages with incorporation of stevia as a natural sweetener. The developed beverages contained 74% whey, 20% amla juice, 6% basil extract, 6% mint, 6% ginger, 6% Alovera and 6% lemon grass that scored the highest acceptability.

MATERIALS AND METHODS

Extraction of herbs

The fresh, matured and cleaned brahmi leaves were collected. 10 g of brahmi leaves was extracted with using 100 ml of distilled water. The fresh, matured and cleaned mint leaves were collected. 10 g of mint leaves was extracted with using 100 ml of distilled water. The cleaned jaljeera powder was weighed as per standardized amount. The extracted herbal extracts except jaljeera powder bottled in glass for further used.

Preparation of whey from milk

The whey was prepared by the method given by Kanchana et al. (2020). The fresh milk was heated upto 90°C for 5-10 min in a stainless steel vessel with continuous stirring by using an electrical stirrer. The heated milk was cooled down to $82-85^{\circ}\text{C}$. The optimized coagulant (0.13% acetic acid) was added slowly into the milk at the temperature of 82°C with slow stirring until the coagulation was done. It was kept upto 10 minutes without any disturbance upto the complete coagulation. The liquid whey separated from the precipitated casein by filtration through pre-sterile muslin cloth. The coagulated whey was pasteurized at $80-85^{\circ}\text{C}$ for 5 mins by using the sterilized vessel. The pasteurized whey was hot-filled in sterile polyethylene terephthalate (PET) and

stored at ($4\pm 1^{\circ}\text{C}$) for further analysis upto 12 h. The protocols for obtaining whey from milk are presented in Figure 1.

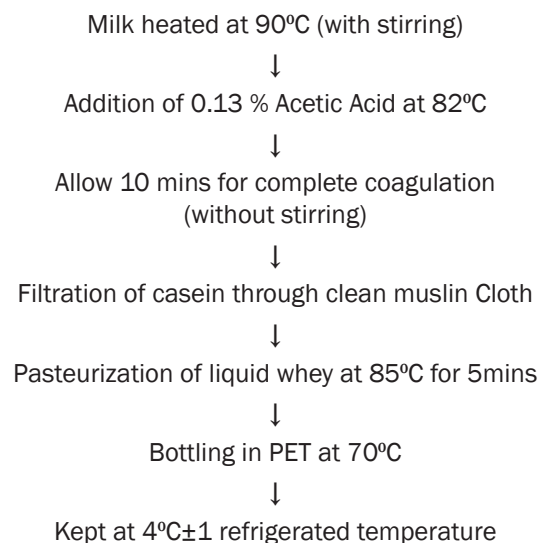


Figure.1. Flow chart for preparation of whey-based herbal drinks

Development of whey-herbal beverages

Whey-based herbal drinks were formulated using whey, sugar, herbal extracts (brahmi, mint and jaljeera), citric acid, pectin, sodium chloride, sodium citrate and water. The ingredients were added into whey and heated at 65°C and pasteurized at $80-85^{\circ}\text{C}$. The protocols for developing whey herbal beverages are presented in Figure 2.

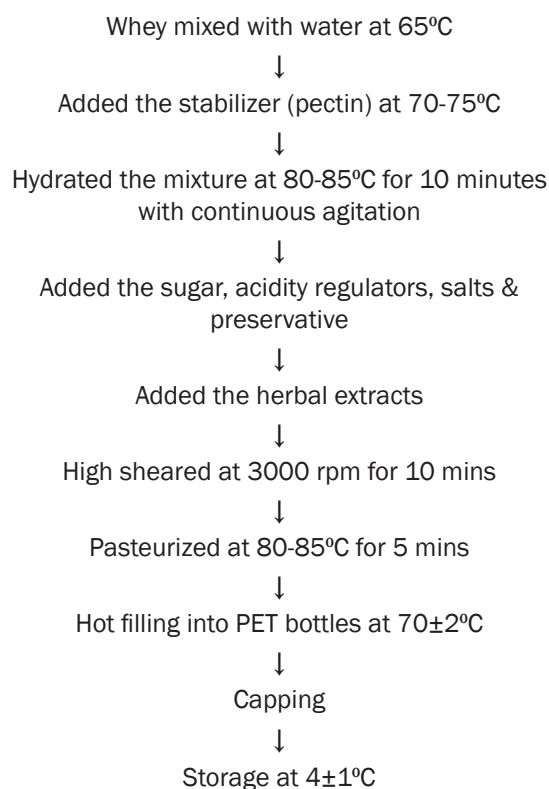


Figure.2. Flow chart for preparation of whey-based herbal drinks

Packaging and storage

The formulated whey-based herbal beverages were hot-filled in the polyethylene terephthalate (PET) and glass bottles and stored at both room temperature ($30\pm 1^\circ\text{C}$) and refrigerated temperature ($4\pm 1^\circ\text{C}$). The sensory attributes of formulated drinks were analyzed in 10 days intervals during 30 days of storage period.

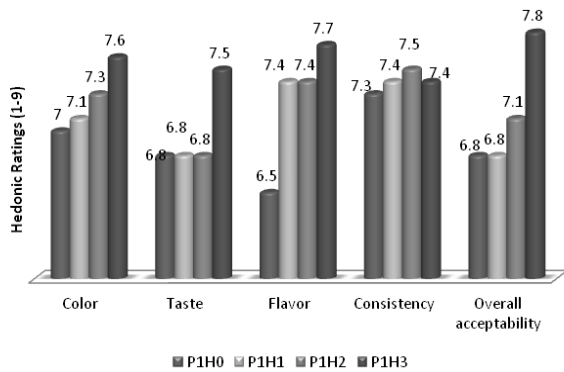


Figure.3. Whey-based herbal drinks packed in PET and stored at room temperature

Physical, chemical and sensory evaluation

The viscosity of the sample was measured by using the method of (Bhuvaneswari and Tiwari, 2007). The color value was determined using the method given by Chauhan *et al.*, (2019). The total solid content was determined by using the Gravimetric method given by FSSAI (2016). The moisture content was determined by the vacuum oven method given by FSSAI (2015). The pH, titrable acidity, protein and fat were estimated by the procedure given in FSSAI (2016). The TSS ($^\circ\text{Brix}$) content was determined by using the Digital Refractometer (Niveadhitha *et al.*, 2018). The lactose, ascorbic acid and calcium content of samples were estimated using the procedure given by Sharma *et al.* (2013). The total sugar, total reducing sugar and total phenol content were analyzed for the formulated drinks (Sadasivam and Manickam, 2008). The total ash content was measured by using the method of Horwitz

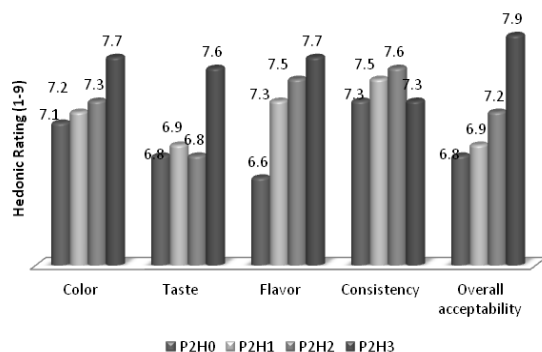


Figure.4. Whey-based herbal drinks packed in glass and stored at room temperature

(1975). The calcium and magnesium content was determined by the procedure given by Sharma *et al.* (2013). The potassium content was estimated by standard procedure and sodium content was estimated by using the flame photometric method given by Ranganna (2000). The sensory evaluation of formulated drinks was done by 9 points hedonic scale rating (Pandey *et al.*, 2019).

Statistical analysis

AGRES Software is applied for analyzing the data collected from the experiments. The statistical analysis was conducted to interpreting the data, find out the stability of formulated drinks and quality of microbial study during the period of storage studies.

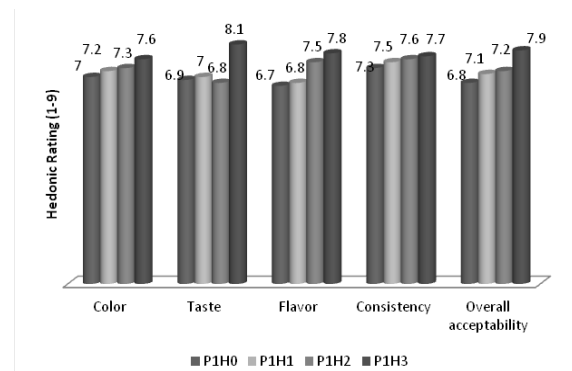


Figure.5. Whey-based herbal drinks packed in PET and stored at refrigerated temperature

RESULTS AND DISCUSSION

Physico-chemical parameters of whey

The pH, titrable acidity (lactic acid) and TSS ($^\circ\text{Brix}$) of obtained whey was 5.21, 0.23% and 6.40, respectively. The viscosity (cp) and color ($L^*a^*b^*$) of whey were 65.8 and 65.8, 0.40, 1.49. The total solid content (8.8 g/100 g) and moisture content (91.2 %) were estimated. The lactose (g/100g), fat (g/100g) and protein (g/100g) of whey were estimated such as 4.288, 0.13 and 0.289, respectively. The pH, titrable acidity (%), total solids (%), fat (%), lactose (%), protein (%) and total ash (%) of 5.0-5.8, 0.2-0.4, 6.5, 0.5, 5.0, 0.4 and 0.5 in medium acid whey reported by Gupta and Prakash (2017). The total ash content was found to be 0.389 g/100 g. The calcium (mg/100g), magnesium (mg/100g), sodium (mg/100g) and potassium (mg/100g) content of whey were estimated 60.12, 10.1, 50.01 and 150.21 respectively. The calcium (mg/100g), magnesium (mg/100g) and sodium (mg/100g) of 92.8, 10 and 30 in acid whey had also been reported by Gupta and Prakash (2017). The physico-chemical parameters of whey are given in Table 1.

Table.1. Physico-chemical parameters of whey

S.No.	Parameters	Mean ± S.D
1.	pH	5.21 0.08
2.	Titration acidity (% Lactic acid)	0.23 0.00
3.	Total soluble solids (°Brix)	6.40 0.09
4.	Viscosity (cp)	2.8 0.04
5.	Color – L*	65.8 0.03
	a*	0.40 0.12
	b*	1.49 0.03
6.	Total solids (g/100g)	8.8 0.10
7.	Moisture (%)	91.2 0.48
8.	Lactose (g/100g)	4.288 0.07
9.	Protein (g/100g)	0.289 0.00
10.	Fat (g/100g)	0.13 0.00
11.	Total ash (g/100g)	0.3892 0.00
12.	Calcium (mg/100g)	60.12 0.74
13.	Magnesium (mg/100g)	10.1 0.27
14.	Sodium (mg/100g)	50.01 0.44
15.	Potassium (mg/100g)	150.21 0.74

Values reported are mean± SD of four replicate

Standardization of ingredients used for whey-based herbal beverages

The whey (65%) was standardized for formulating whey based herbal beverages. The three herbs (brahmi, mint and jaljeera powder) were selected for the study. The brahmi extract (3%), mint (2%) and jaljeera powder (0.15%) were standardized. The ingredients used for whey-based herbal beverages is given in Table 2. Using the ingredients and standardized protocol given in Figure 2, the whey-based herbal beverages were formulated.

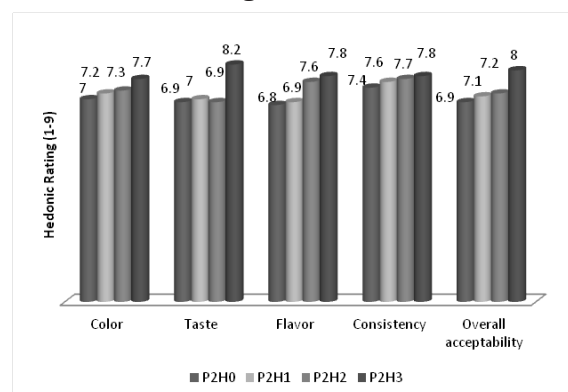


Figure.6. Whey-based herbal drinks packed in glass and stored at refrigerated temperature

Table.2. Ingredients used for whey-based herbal beverages

Ingredients (%)	Treatments			
	H ₀	H ₁	H ₂	H ₃
Whey	65	65	65	65
Sugar	11	11	11	11
Citric acid	0.01	0.01	0.01	0.01
Sodium citrate	0.02	0.02	0.02	0.02
Pectin	0.02	0.02	0.02	0.02
Salt	0.025	0.025	0.025	0.025
Brahmi extract	-	3.00	-	-
Mint extract	-	-	2.00	-
Jaljeera powder	-	-	-	0.15
Water	23.93	20.93	21.93	23.77

H₀ : 65% whey + without herbal (control) ; H₁ : 65% whey + 3% brahmi extract ; H₂ : 65%

whey + 2% mint extract ; H₃ : 65% whey + 0.15% jaljeera powder

Physical characteristic of whey-herbal drinks

The physical characteristics of whey-based herbal drinks are given in Table 3. The viscosity (cp) of the whey-herbal drinks (H₀, H₁, H₂ and H₃) was found to be 3.2, 3.3, 3.3 and 3.4, respectively. The L*, a*, b* value for H₀ were 57.2, 0.49, -1.54 for H₁ were 35.71, 30.71, -0.43 for H₂ were 32.42, 35.32, -0.56 and for H₃ were 20.2, 1.65, -1.54 respectively. Nearly similar findings of color value (L*) 20.67, (a*) -3.73, (b*) 6.5 in mint incorporated lemonade beverages were reported by Tamer *et al.* (2017). Hemalatha *et al.* (2018) reported that the viscosity (cp) of Cape gooseberry RTS beverages were 28.73. The reason

Table.3. Physical characteristic of whey based herbal beverages

S.No. Treatments	Color			Viscosity
	L*	a*	b*	
1. H ₀	57.2	0.49	-1.54	3.2
2. H ₁	35.71	30.71	-0.43	3.3
3. H ₂	32.42	35.32	-0.56	3.3
4. H ₃	20.2	1.65	-1.54	3.4
Statistical values	Color			Viscosity
	L*	a*	b*	
SED	0.1973	0.3159	0.0201	0.0394
CD (0.01)	0.6028	0.9648	0.0615	0.1205
CD (0.05)	0.4300**	0.6882**	0.0439**	0.0859**

H₀ : 65% whey + without herbal (control) ; H₁ : 65% whey + 3% brahmi extract ; H₂ : 65% whey + 2% mint extract ; H₃ : 65% whey + 0.15% jaljeera powder

for increasing the viscosity of the RTS beverage might have included a stabilizer to the beverage. There was a significant difference on viscosity and color (L^* , a^* , b^* value) were observed between the formulated whey based herbal drinks.

Chemical characteristic of whey-herbal drinks

The chemical characteristics of whey-based herbal drinks are given in Table 4. There was a significant difference in pH were found between the herbal drinks, and H_3 (65% whey: 0.15% jaljeera) had higher pH (5.24) compared to H_0 , H_1 and H_2 . Baljeet et al.(2013) reported that the addition of herbal extract increased the pH of the drink. The titrable acidity (% citric acid) of H_0 (0.25), H_1 (0.20), H_2 (0.23) and H_3 (0.21) significantly differed among all whey herbal drinks. The result of titrable acidity was on par with results reported by Sharma et al.(2019). H_3 recorded the highest TSS content (12°Brix). Apart from added sugar to drink, sugar content of jaljeera powder is contributed to the increased TSS content of H_3 . The lactose content (2.78g/100g) was not significantly differed between the herbal drinks (H_0 , H_1 , H_2 and H_3). The results were nearin agreement with the findings reported by (Sharma et al., 2019). The protein, fat, total reducing sugar and total sugar

content of H_3 were found to be highest 0.46g/100g, 0.30g/100g, 3.43g/100g and 14.43g/100g than H_0 , H_1 and H_2 . And there was a significant difference on protein, fat, total reducing sugar and total sugar were observed between the all herbal drinks. The result of fat content was agreed with Chavanet al.(2015a). Baljeet et al.(2013) reported results of total sugar were on par with total sugar content of formulated whey-herbal drinks. The total ash (g/100g) and total phenol (mgGAE/100g) content of was increased in H_2 were found to be 0.43 and 219.96, respectively. The ascorbic acid (mg/100g) content was highest in H_1 (1.89) and calcium (mg/100g) content was highest in H_3 (55.27). And there was a significant difference in total ash, calcium and ascorbic acid were observed between all herbal drinks. Baljeet et al.(2013) reported that the ascorbic acid of 0.2% mint incorporated whey-based pineapple-bottle gourd beverage were 1.42 mg/100g.Sharma et al.(2019) reported that the calcium content of whey-apple herbal beverage (jaljeera) was15.68 mg/100g. The highest energy value (kcal/100g) was found in H_3 (62.26). There was a significant difference in energy value (kcal/100g) in the formulated whey-herbal drinks (H_0 , H_1 , H_2 and H_3).

Table.4. Chemical characteristics of whey based herbal drinks

Parameters	H_0	H_1	H_2	H_3	SED
pH	5.10	5.25	5.21	5.24	0.0429*
Titrable acidity (citric acid %)	0.25	0.20	0.23	0.21	0.0027**
TSS ($^{\circ}$ Brix)	11.6	11.9	11.8	12.0	0.1634*
Moisture (%)	85.62	85.46	85.04	84.42	0.0234 ^{NS}
Lactose (g/100g)	2.78	2.78	2.78	2.78	0.0521 ^{NS}
Protein (g/100g)	0.18	0.25	0.28	0.46	0.0030**
Fat (g/100g)	0.08	0.11	0.16	0.30	0.0016**
Total Ash (g/100g)	0.25	0.31	0.43	3.43	0.0027**
Total reducing sugar (g/100g)	2.80	3.11	2.99	14.43	0.1409**
Total sugar (g/100g)	13.81	14.11	13.99	55.27	0.1301**
Calcium (mg/100g)	39.07	45.14	39.07	0.12	0.7427**
Ascorbic acid (mg/100g)	ND	1.89	0.64	14.26	0.0096**
Total phenol (mgGAE/100g)	ND	138.72	219.96	62.26	1.8041**
Energy (kcal/100g)	56.68	58.44	58.52	62.26	1.0342**

H_0 : control (65% whey without herbal); H_1 : 65% whey + 3% Brahmi extract ; H_2 : 65% whey + 2% mint extract ; H_3 : 65% whey + 0.15% jaljeera powder, ND-Not detected, NS-Non Significant, **-highly significant.

Sensory evaluation

The sensory attributes such as color, taste, flavor, consistency and overall acceptability of formulated whey-based herbal beverages were evaluated by hedonic 9 point scale. There was a significant difference on sensory attributes were observed

between the formulated herbal beverages. The whey-based jaljeera drink (H_3) had scored highest color (7.84/9.0), taste (8.32/9.0), flavor (8.02/9.0), consistency (7.98/9.0) and overall acceptability (8.10).The sensory evaluation of formulated whey beverages aregiven in Table 5.

Table.5. Sensory evaluation of whey-based herbal drinks

Treatments	Color	Taste	Flavor	Consistency	Overall acceptability
H ₀	7.10	7.00	6.98	7.50	7.05
H ₁	7.35	7.15	7.01	7.75	7.25
H ₂	7.40	7.04	7.78	7.85	7.45
H ₃	7.84	8.32	8.02	7.98	8.10
SED	0.1064	0.0956	0.1036	0.1187	0.0734
CD (0.01)	0.3251	0.2921	0.3166	0.3625	0.2241
CD (0.05)	0.2319**	0.2083**	0.2258**	0.2586**	0.1598**

H₀ : 65% whey + without herbal (control) ; H₁ : 65% whey + 3% brahmi extract ; H₂ : 65% whey + 2% mint extract ; H₃ : 65% whey + 0.15% jaljeera powder, **-Highly significant

Changes in sensory attributes during storage

There was a significant difference ($p < 0.05$) on sensory attributes such as color, taste, flavor, consistency and overall acceptability were found between the H₀, H₁, H₂ and H₃ (Figure 3 to Figure 6). The sensory attributes of the whey-herbal drinks were significantly ($p < 0.05$) decreased with respect to increasing storage period. There was a significant difference found between the packaging materials (PET and glass). The beverages that were bottled in glass (P₂) were retained more sensory attributes than beverages stored at PET (P₁). There was a significant difference found between the storage conditions (room temperature and refrigerated temperature). The beverages were stored in room temperature (R₁) were retained more sensory attributes than beverages stored at refrigerated temperature (R₂). The results of sensory evaluation of formulated whey-herbal beverages bottled in glass scored the highest acceptability compared to packed in PET. The decreasing sensory attributes were also reported by Baljeet *et al.* (2013). Similar results were found in whey-pineapple-based bottle



H₀ : 65% whey + without herbal (control) ; H₁ : 65% whey + 3% brahmi extract ; H₂ : 65% whey + 2% mint extract ; H₃ : 65% whey + 0.15% jaljeera powder

Figure.7. Whey-based herbal drinks packed in PET (P₁)

gourd herbal beverage during storage reported by Baljeet *et al.* (2013).



H₀ : 65% whey + without herbal (control) ; H₁ : 65% whey + 3% brahmi extract ; H₂ : 65% whey + 2% mint extract ; H₃ : 65% whey + 0.15% jaljeera powder

Figure.8. Whey-based herbal drinks packed in glass (P₂)

CONCLUSION

The incorporation of herbal extracts to the whey was more suitable for converting the whey (by-product) into value-added products. The fat content of the formulated whey-based herbal beverages is less than 1% so it may be considered as nearly fat-free healthy drinks. The pasteurized whey-herbal drinks were bottled in glass and stored at $4 \pm 1^\circ\text{C}$ was retained more sensory attributes compared to packed in polyethylene terephthalate (PET) and stored at $30 \pm 1^\circ\text{C}$. Based on sensory studies conducted, the acceptable consumption period for whey-based herbal drinks were 10 days and 20 days under $30 \pm 1^\circ\text{C}$ and $4 \pm 1^\circ\text{C}$, respectively. The cost of the formulated whey-based herbal beverages between Rs.15-20/250mL was less than the commercially available other herbal beverages. The formulated whey beverage had less fat and low calorie, which may be recommended for weight-reducing people.

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