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Comparative study on qualitative analysis of various phytochemicals in herbal yoghurt at different storage periods

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Abstract

Yoghurt is 'the coagulated milk product obtained by lactic acid fermentation through the action of *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus salivarius* subsp. *thermophilus*' The beneficial health effects of yoghurt have been partly linked to the proteolysis products produced during fermentation and storage. The present study investigate infusion of the commonly used herbs *Aloe barbadensis* (*Aloe vera*), *Anethum graveolens* (dill), *Costus igneus* (insulin plant) and *Hibiscus sabdariffa* (roselle) on yoghurt formation and also to evaluate the presence of various phytochemicals in detail at different storage periods. It was observed from the study that phytochemicals were detected in the all yoghurts during storage period of 14 days and no phytochemicals were noticed on 21st day of storage.

Keywords: Herbal, yoghurt, storage period, qualitative analysis, phytochemicals

Introduction

Yoghurt is 'the coagulated milk product obtained by lactic acid fermentation through the action of *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus salivarius* subsp. *thermophilus*' The beneficial health effects of yoghurt have been partly linked to the proteolysis products produced during fermentation and storage. During fermentation, LAB produces a range of secondary metabolites, some of which have been associated with health promoting properties of which the notable ones are the B-vitamins and bioactive peptides. Yoghurt is a widely consumed functional food due to its good taste and nutritional properties.

These are secondary metabolites of plant origin that constitute one of the most abundant groups of natural metabolites and form an important part of both human and animal diets ^[14]. This opens the possibility that consumption of select phenolic phytochemicals rich foods may mimic synthetic inhibitors and provide health benefits, but without adverse side effects.

The present study investigate infusion of the commonly used herbs *Aloe barbadensis* (*Aloe vera*), *Anethum graveolens* (dill), *Costus igneus* (insulin plant) and *Hibiscus sabdariffa* (roselle) on yoghurt formation and also to evaluate the presence of various phytochemicals in detail at different storage periods..

Materials and methods

Fresh cow milk obtained from the Department of ILFC, Veterinary College and Research Institute, Namakkal – 637 002 was used. Skim milk powder testing 5 and 95 % moisture and solubility, respectively was purchased locally (AAVIN). Commercially available good quality canesugar was used in the current study. Freeze dried DVS cultures containing yoghurt bacteria *Lactobacillus delbrueckii* ssp. *bulgaricus* and *Streptococcus salivarius* ssp. *thermophilus* obtained from Chr. Hansen, Denmark, were used in this study. Herbal infusions like *Aloe barbadensis* collected from campus of Veterinary College and Research Institute, Namakkal. *Anethum graveolens* purchased from local markets of Kolar district, Karnataka. *Costus igneus* and *Hibiscus Sabdariffa* collected from Horticulture College, GKVK campus, Bangalore was employed in the study. The different herbal plants used in the current study were depicted as figure 1 to 4.



Fig 1: *Anethum graveolens*



Fig 2: *Aloe barbadensis*



Fig 3: *Costus igneus*



Fig 4: *Hibiscus sabdariffa*

Preparation of probiotic yoghurt

Six batches of different herbal probiotic yoghurt were prepared using fresh milk. Skim milk powder at the rate of 2 % (w/v) and sugar at the rate of 6 % (w/v) were added to it and homogenized at 1000 psi. The contents were mixed well and pasteurized at 85°C for 30 minutes, cooled to room temperature and inoculated with 4 % of yoghurt cultures

containing *Lactobacillus delbrueckii* subsp. *bulgaricus*, and *Streptococcus salivarius* subsp. *thermophilus*. Different concentrations of herbs were added before incubation and mixed well and incubated at 42°C for 4 to 5 hours and stored at 4°C when the pH was reduced to 4.5. These yoghurts were then placed in the refrigerator for up to 21 days.

Herbal yoghurt

Five different concentrations of herbal yoghurt were prepared from each different herb for standardization and were denoted as

- PY = Plain yoghurt without adding any herb as a standard yoghurt
- ABY = *Aloe barbadensis* incorporated yoghurt
- AGY = *Anethum graveolens* incorporated yoghurt
- CIY = *Costus igneus* incorporated yoghurt
- HSY = *Hibiscus sabdariffa* incorporated yoghurt

Different concentrations used for standardization of herbal yoghu

Thus prepared herbal yoghurt was subjected to evaluate the presence of various phytochemicals in detail at different storage periods.

Yoghurt	Concentration of herbs (per cent)				
PY	0	0	0	0	0
ABY	12	14	16	18	20
AGY	0.4	0.5	0.6	0.7	0.8
CIY	0.4	0.5	0.6	0.7	0.8
HSY	0.4	0.5	0.6	0.7	0.8

Storage period used in the current study

Herbal yoghurt was subjected to evaluate the presence of various phytochemicals in detail at different storage periods like zero, 7th, 14th and 21st day.

Results

The outcome of the present study on Qualitative analysis of phytochemicals in herbal yoghurt is depicted as Tables 1, 2, 3, 4 and 5 show the qualitative analysis of phytochemicals in PY, ABY, AGY, CIY and HSY during storage period of zero to 14 days, which were as follows:

Phytochemicals	PY			
	Zero day	7 th day	14 th day	21 st day
Flavanoids	-	-	-	-
Saponin	-	-	-	-
Terpenoids	+	+	+	-
Tannin	-	-	-	-
Glycosides	+	+	+	-
Steroids	-	-	-	-
Alkaloids	-	-	-	-

Table 1: Qualitative analysis of phytochemicals in plain yoghurt

Phytochemicals	ABY			
	Zero day	7 th day	14 th day	21 st day
Flavanoids	+	+	+	-
Saponin	-	-	-	-
Terpenoids	+	+	+	-
Tannin	-	-	-	-
Glycosides	+	+	+	-
Steroids	+	+	+	-
Alkaloids	-	-	-	-

Table 2: Qualitative analysis of phytochemicals in *Aloe barbadensis* yoghurt

Phytochemicals	AGY			
	Zero day	7 th day	14 th day	21 st day
Flavanoids	+	+	+	-
Saponin	+	+	+	-
Terpenoids	+	+	+	-
Tannin	+	+	+	-
Glycosides	+	+	+	-
Steroids	+	+	+	-
Alkaloids	-	-	-	-

Table 3: Qualitative analysis of phytochemicals in *Anethum graveolens* yoghurt

Phytochemicals	CIY			
	Zero day	7 th day	14 th day	21 st day
Flavanoids	+	+	+	-
Saponin	-	-	-	-
Terpenoids	+	+	+	-
Tannin	+	+	+	-
Glycosides	+	+	+	-
Steroids	-	-	-	-
Alkaloids	+	+	+	-

Table 4: Qualitative analysis of phytochemicals in *Costus igneus* yoghurt

Phytochemicals	HSY			
	Zero day	7 th day	14 th day	21 st day
Flavanoids	+	+	+	-
Saponin	-	-	-	-
Terpenoids	+	+	+	-
Tannin	+	+	+	-
Glycosides	+	+	+	-
Steroids	-	-	-	-
Alkaloids	-	-	-	-

Table 5: Qualitative analysis of phytochemicals in *Hibiscus sabdariffa* yoghurt

PY:	Terpenoids and glycosides;
ABY:	Flavanoids, terpenoids, glycosides and steroids;
AGY:	Flavanoids, saponin, terpenoids, tannin, glycosides and steroids;
CIY:	Flavanoids, tannin and alkaloids, terpenoids and glycosides;
HSY:	Flavanoids, tannin, terpenoids and glycosides

It was observed from the tables that the phytochemicals were present in the all yoghurts during storage period of 14 days which disappeared on day 21 of storage.

Discussion

Varieties of naturally-occurring minor components (chemicals or substances) present in plants [2] are called 'phytochemicals' [10]. These include flavonoids, tocopherols, carotenoids, and ascorbic acids [7]. Various types of phytochemicals observed in the herbal yoghurt are as follows

PY: Terpenoids and glycosides;

ABY: Flavanoids, terpenoids, glycosides and steroids;

AGY: Flavanoids, saponin, terpenoids, tannin, glycosides and steroids;

CIY: Flavanoids, tannin and alkaloids, terpenoids and glycosides;

HSY: Flavanoids, tannin, terpenoids and glycosides

It was observed from the study that phytochemicals were detected in the all yoghurts during storage period of 14 days and no phytochemicals were noticed on 21st day of storage.

These results are in accordance with Sathyaprabha *et al.* (2010) who recorded the flavonoid, terpenoids, Saponin and Tannin in *Aloe vera* gel. Kumar *et al.*, (2012) reported the presence of flavonoids, terpenoids, tannin and glycosides in aqueous extract of *H. sabdariffa*. Pazhanichamy (2012) noticed the presence of flavonoids, tannin and alkaloids in aqueous extract of *C. igneus*. Jana and Shekhawat (2010) found flavonoids, tannin and alkaloids in aqueous extract of *A. graveolens*.

The presence of glycosides and terpenoids in the plain yoghurt may be due to the reason that glycosides and terpenoids are compounds containing a carbohydrate and non-carbohydrate residue in the same molecule and these are present in the animal constituents like milk and tissues [6], and on addition of concentrated H₂SO₄ formation of reddish brown colour at the interface mimic presence of glycosides and terpenoids.

The absence of phytochemicals in 21st day of storage period is may be due to the following reasons [1].

The stability of phytochemicals in food products during storage period were most affected by water activity, temperature, light, enzymatic oxidation and pH.

Polyphenols are readily oxidised during storage and which results in the production of H₂O₂.

Conclusion

The phytochemicals were detected in all herbal extract supplemented yoghurt during storage period of 14 days and no phytochemicals were detected on 21st day of storage.

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