Full Length Article

ANTIOXIDANT ACTIVITY OF HERBAL YOGHURT INCORPORATED WITH CINNAMON (*CINNAMOMUM ZEYLANICUM*) EXTRACT

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ABSTRACT

The aim of the study was to develop herbal yoghurt by incorporating cinnamon bark extract (Cinnamomum zeylanicum) at 0.6 and 1.2 per cent level. The developed herbal yoghurt samples were analysed for physico-chemical properties, antioxidant activity and sensory quality by standard procedures. The incorporation of cinnamon extract improved the antioxidant activity of yoghurt significantly. The antioxidant activity of control and cinnamon incorporated yoghurt (0.6 and 1.2 per cent) were 28.12±2.16, 44.87±2.16 and 54.77±2.16 per cent, respectively. A decline in antioxidant activity was observed in all yoghurt samples after the third day of storage. No significant difference in fat and total solids content was observed in yoghurt samples incorporated with cinnamon extract. The sensory scores of yoghurt incorporated with 0.6 per cent cinnamon extract were significantly higher than that of control. The study revealed that cinnamon extract can be incorporated into yoghurt to enhance the antioxidant activity without compromising the sensory quality.

Keywords: Yoghurt, cinnamon, antioxidant activity, herbal yoghurt

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INTRODUCTION

Yoghurt is a nutritious dairy product obtained from milk by bacterial fermentation. It is the most widely consumed fermented dairy product throughout the world. It acts as an ideal vehicle for functional ingredients. The fermentation of the yoghurt is by the action of two organisms, viz. *Streptococcus thermophilus* and *Lactobacillus delbrueckii ssp. bulgaricus.* These two cultures are generally added in a ratio of 1:1. The fermentation of yoghurt begins with the breakdown of lactose into glucose and galactose. The basic structure and texture of yoghurt are formed by lactic acid, whereas other molecules contribute to the flavour of yoghurt.

In recent years, food producers have the tendency to incorporate herbal ingredients to improve the nutritional and functional properties of foods

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(Illupapalayam *et al.*, 2014). These herbals also help to extend the shelf life by retarding the growth of food-borne pathogens. The incorporation of herbs and spices or their extracts into different milk products makes the products carriers of nutraceuticals (El-Sayed and Youssef, 2019). Various herbs such as clove (*Syzygium aromaticum*), thyme (*Thymus vulgares*), rosemary (*Rosemarinus officinalis*), turmeric (*Curcuma longa*) and cinnamon (*Cinnamomum zeylanicum*) are added to improve the nutritional as well as functional properties of yoghurt (Paswan *et al*., 2021).

Cinnamomum zeylanicum (Cinnamon) is an evergreen short tree belonging to the family Lauraceae and it is common in South India and Sri Lanka. Cinnamon bark is widely used as a spice in many food stuffs. In addition to the flavouring properties, they have various health benefits. The essential oils of cinnamon have antioxidant, antidiabetic, antifungal and antimicrobial activity. Cinnamon showed antibacterial activity against food-borne pathogens such as *S. typhi, E. coli, S. aureus, etc.* (Muhammad and Dewettinck, 2017). These beneficial characteristics can be used to improve food safety and to enhance the shelf life of fat rich dairy and food products.

MATERIALS AND METHODS

Preparation of aqueous extract of *Cinnamomum zeylanicum* bark

Modified procedure of Behard *et al*. (2009) was used for the preparation of cinnamon bark extract. The dried cinnamon bark was semi-grounded by using an electric blender (Bajaj, Ambala, India). A hundred grams of powder was then wrapped in a muslin cloth and immersed in a beaker containing autoclaved distilled water at 60°C. The beaker was then kept in a water bath maintained at 70°C till the complete extraction occurred. Then the extract was kept in a rotatory vacuum evaporator (Buchi) at 45°C under reduced pressure.

Lyophilization of Cinnamon extract

After the evaporation of excess water from the extract by using a rotary flash evaporator, the concentrated extract of *C. zeylanicum* was freeze dried in a lyophilizer (Operon, -70°C freeze dryer). The lyophilized extract was then stored at refrigerated temperature for further use.

Preparation of yoghurt

Yoghurt was prepared as per the method of Tamime and Robinson (2007). Cow milk was used for the preparation of yoghurt after adding three per cent skim milk powder. It was then heated at 90°C for 15 minutes and six per cent sugar was added. Then the contents were mixed properly and cooled to 42°C. Two per cent yoghurt culture (NCDC-145) obtained from the National Collection of Dairy Culture (NCDC), Karnal was added. Two experimental yoghurt samples were prepared by adding 0.6, 1.2 per cent aqueous extract of *C. zeylanicum*. It was then incubated at 37°C overnight.

Physico – chemical analysis of functional yoghurt

The titratable acidity of yoghurt samples was determined according to the procedure laid out in the FSSAI (2015). The fat content of yoghurt was determined by Rose Gottlieb method as per FSSAI (2015). The total solids content of yoghurt samples was determined according to the procedure described by the Bureau of Indian Standards (IS 12333, 1997). The syneresis percentage was determined as per the procedure described by Doleyres and Lacroix (2005). Syneresis was expressed as the percentage weight of drained whey over the initial weight of the yoghurt sample.

The antioxidant activity of yoghurt samples was determined by the ability of each extract to scavenge 1, 1- diphenyl - 2 - picryl hydrazyl (DPPH) radicals by using the procedure laid out by Debasree Ghosh (2019) with slight modifications. The antioxidant activity of yoghurt is expressed as the inhibition percentage of DPPH free radicals.

Sensory evaluation

Sensory evaluation of yoghurt samples was carried out by faculty members and postgraduate students of the Department of Dairy Science. Yoghurt samples were evaluated for their sensory characteristics such as colour and appearance, body and texture, flavour and overall acceptability as per the scorecard suggested by IDF (1987).

RESULTS AND DISCUSSION

Titratable acidity

The mean titratable acidity values of control yoghurt (C) were 0.96±0.01, 1.04±0.01, 1.15±0.02 and 1.22±0.02 per cent on the first, third, fifth and seventh days of storage, respectively. The respective mean values of yoghurt containing 0.6 per cent cinnamon extract (T1) were 0.97±0.01, 1.02±0.01, 1.13±0.02 and 1.19±0.02 per cent. The corresponding values of yoghurt containing 1.2 per cent of cinnamon extract (T2) were 0.98±0.01, 1.0±0.01, 1.11±0.02 and 1.16±0.02 percentage, respectively. No significant ($p>0.05$) difference in titratable acidity was observed between the control and treatment groups. However, during storage a significant ($p<0.05$) increase in titratable acidity was shown by control and treatment groups of yoghurt samples. Higher titratable acidity values were observed in control yoghurt, which was significantly higher than T2. The titratable acidity of T1 and control were not significantly ($p>0.05$) different. According to Food Safety and Standards Regulations (2011), yoghurt should have a minimum titratable acidity value of 0.6 per cent lactic acid. The incorporation of 1.5 per cent cinnamon (*Cinnamomum cassia* and *Cinnamomum verum*) had extended the fermentation time up to 22 hours and cinnamon had clearly inhibited the growth of lactic acid bacteria (Nabavi *et al.*, 2015). Suliman *et al.* (2019) and Pavalakumar *et al.* (2021) also observed the concentration of cinnamon and storage period had a significant impact on the titratable acidity of yoghurt. Titratable

acidity decreased slightly during storage as cinnamon concentration increased. The low acidity values in yoghurt incorporated with cinnamon could be attributed to the presence of antibacterial components in cinnamon, such as cinnamaldehyde and eugenol which could have suppressed the growth of starter cultures.

Fat percentage

The mean fat content of control yoghurt (C) on the first, third, fifth and seventh days of storage were 3.35±0.06, 3.34±0.07, 3.32±0.07 and 3.30±0.07 per cent respectively. The corresponding values of yoghurt with 0.6 per cent cinnamon bark extract (T1) were 3.35±0.06, 3.35±0.07, 3.34±0.07 and 3.34±0.07 per cent. For yoghurt with 1.2 per cent cinnamon extract (T2), the values were 3.36±0.06, 3.36±0.07, 3.35±0.07 and 3.35±0.07 per cent respectively. There was no significant ($p>0.05$) difference in fat content between control and treatment groups of yoghurt samples. The results are in agreement with the findings of Vidanagamage *et al.* (2016) and Gunes-Bayir and Bilgin (2019). They reported that cinnamon incorporated butter showed no significant difference in fat percentage. However, Adam Mohamed (2018) found that the fat content of cinnamon-added yoghurt increased with increasing concentrations of cinnamon.

Total solids content

The mean total solids per cent of control yoghurt samples (C) were 20.77±0.69, 21.65±0.24, 21.94±0.33 and 21.74±0.47 on the first, third, fifth and seventh days of storage respectively. The respective values for yoghurt incorporated with 0.6 per cent cinnamon extract (T1) were 21.22±0.69, 22.19±0.24, 22.32±0.33 and 22.45±0.47 per cent. The values for yoghurt incorporated with 1.2 per cent cinnamon bark extract (T2) were 21.23±0.69, 22.27±0.24, 22.26±0.33 and 22.07±0.47 per cent, respectively. No significant ($p>0.05$) difference in total solids per cent was observed between control and treatment groups of yoghurt samples. However, during storage total solids content showed a significant increase ($p<0.05$) on fifth and seventh days when compared to first day. Higher total solids content was shown by T1 but it was not significantly different from T2 and control. These results are in accordance with the findings of Khadka (2018) and Sanjay *et al*. (2020). The increase in total solids might be due to the presence of crude fibre and mineral content present in cinnamon. During storage, a significant increase in total solids content was observed and it could be attributed to the evaporation of moisture during refrigerated storage.

Syneresis percentage

The mean syneresis values of control yoghurt (C) on the first, third, fifth and seventh days were 3.04±0.16, 2.77±0.27, 2.73±0.14 and 2.78±0.16 per cent respectively. The corresponding values for yoghurt incorporated with 0.6 per cent cinnamon bark extract (T1) were 2.99±0.16, 2.85±0.27, 3.02±0.14 and 2.91±0.16 per cent. The values for yoghurt with 1.2 per cent cinnamon extract (T2) were 3.05±0.16, 2.64±0.27, 3.06±0.14 and 3.20±0.16 per

cent respectively. No significant ($p>0.05$) difference in syneresis percentage was observed between control and treatment groups of yoghurt samples. However, during storage, there was a significant ($p<0.05$) decrease in syneresis on fifth day of storage in control samples. No significant difference in syneresis values were observed between treatment groups of yoghurt samples during storage. The highest syneresis was observed in yoghurt with 1.2 per cent cinnamon extract (T2) but it was not significantly ($p>0.05$) different from control and 0.6 percent cinnamon extract (T1), Khadka (2018) reported that the syneresis of yoghurt containing cinnamon oleoresin increased from 22.61 to 40.60 per cent during refrigerated storage. Yoghurt added with cinnamon had a higher rate of syneresis than the control sample (Jimenez-Redondo *et al.,* 2022 and Tang *et al.,* 2022). It could be attributed to the presence of polyphenols, which react with proteins and destabilise the casein micelles which could have increased the spontaneous syneresis.

Antioxidant activity

The mean antioxidant activities of control yoghurt (C) on the first, third, fifth and seventh day of storage were 28.12±2.16, 21.77±0.88, 20.55±0.83 and 19.17±1.03 per cent respectively. The mean values of yoghurt incorporated with 0.6 per cent cinnamon bark extract (T1) were 44.87±2.16, 31.65±0.88, 28.82±0.83 and 22.50±1.03 per cent respectively. The corresponding mean values for yoghurt with 1.2 per cent cinnamon extract (T2) were 54.77±2.16, 34.50±0.88, 30.53±0.83 and 23.60±1.03 per cent. Yoghurt incorporated with cinnamon extract had significantly ($p<0.05$) higher antioxidant activity than the control. When the concentration of cinnamon increased, a significant increase in antioxidant activity was observed. Antioxidant activity of treatment groups of yoghurt samples showed a significant reduction ($p<0.05$) from day three to seven. During the same days, control showed a non-significant decrease in antioxidant activity. An increase in antioxidant activity with the addition of cinnamon was also reported by earlier researchers. The antioxidant activity of yoghurt incorporated with cinnamon (six per cent (w/v)) was 31.8 per cent and that of plain yoghurt was 21.8 per cent (Behard *et al.,* 2009). Similar findings were also reported by Tagliazucchi *et al.* (2016) and Helal and Tagliazucchi (2018). Cinnamaldehyde and its derivatives cinnamic acid, methyl cinnamate and cinnamyl alcohol present in cinnamon might have contributed to the higher antioxidant activity. The presence of radical scavenging activity in the control formulations could be attributed to bioactive peptides found in yoghurt. The reduced radical scavenging activity during refrigeration is possibly due to increased interactions between milk proteins and cinnamon polyphenols during storage.

Sensory evaluation

The mean sensory scores of control yoghurt (C) for appearance and colour, body and texture, flavour and overall acceptability were 5.00±0.00, 5.00±0.00, 9.14±0.26 and 19.14±0.26 respectively. The sensory scores

for yoghurt with 0.6 per cent cinnamon extract (T1) were 5.00±0.00, 5.00±0.00, 9.86±0.14 and 19.86±0.14. Scores for yoghurt with 1.2 per cent cinnamon extract (T2) were 5.00±0.00, 5.00±0.00, 9.43±0.30 and 19.43±0.30 respectively. Control and treatment groups showed no significant ($p>0.05$) difference in appearance and colour, body and texture scores of yoghurt samples. A non-significant ($p>0.05$) difference was noticed between the flavour score of control and T2. Flavour score obtained for T1 was significantly ($p<0.05$) higher than T2 and C. Hence the highest overall acceptance score was obtained for T1. Incorporation of cinnamon extract at 0.6 per cent level had the highest consumer acceptance in terms of flavour and overall acceptability. In yoghurt with 1.2 percent cinnamon extract the flavour and overall acceptance scores were higher than that of control but it was not statistically significant. Flavour is normally considered to be the most important and critical indicator of yoghurt quality than other quality parameters like colour, appearance, thickness and texture of yoghurt (Bodyfelt *et al.,* 1988). Yadav and Shukla (2014) reported that yoghurt with one per cent cinnamon received higher scores for flavour and overall acceptance. Similar observation was also reported by Gunes-Bayir and Bilgin (2019). According to Jimenez-Redondo *et al.* (2022), the addition of 0.5 per cent cinnamon did not interfere with the fermentation of lactic acid bacteria and allowed proper fermentation of the yoghurt, resulting in the preservation of yoghurt quality matrices and high customer acceptability scores. However, yoghurt samples with 1.5 per cent cinnamon were not acceptable.

CONCLUSION

The results obtained in the study provide important insight into the production of herbal yoghurt from cow milk by incorporating cinnamon extract. The incorporation of cinnamon extract improved the antioxidant activity of yoghurt significantly. No significant difference in fat and total solids, titratable acidity and syneresis was observed in yoghurt samples incorporated with cinnamon extract. The sensory scores of yoghurt incorporated with 0.6 per cent cinnamon extract were significantly higher than that of control. Hence, the incorporation of plant extracts such as cinnamon would be a better option to improve the antioxidant activity naturally in dairy products

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Table 1. Physico-chemical parameters of yoghurt (Mean ± SE)

Parameters	Samples	Storage Days			
		1st Day	3rd Day	5th Day	7th Day
Titratable acidity (in percent lactic acid)	Control	0.96±0.01^{a}	1.04±0.01^{Ab}	1.15±0.02^{c}	1.22±0.02^{Ad}
	T1(0.6% cinnamon)	0.97±0.01^{a}	1.02±0.01^{ABb}	1.13±0.02^{c}	1.19±0.02^{ABc}
	T2 (1.2% cinnamon)	0.98±0.01^{a}	1.0±0.01^{Bb}	1.11±0.02^{c}	1.16±0.02^{Bc}
Fat (%)	Control	3.35±0.06	3.34±0.07	3.32±0.07	3.30±0.07
	T1(0.6% cinnamon)	3.35±0.06	3.35±0.07	3.34±0.07	3.34±0.07
	T2 (1.2% cinnamon)	3.36±0.06	3.36±0.07	3.35±0.07	3.35±0.07
Total solids content (%)	Control	20.77±0.69^{a}	21.65±0.24^{ab}	21.94±0.33^{b}	21.74±0.47^{b}
	Total solids content (%)	21.22±0.69^{a}	22.19±0.24^{ab}	22.32±0.33^{b}	22.45±0.47^{b}
	T2 (1.2% cinnamon)	21.23±0.69^{a}	22.27±0.24^{ab}	22.26±0.33^{b}	22.07±0.47^{b}
Syneresis (%)	Control	3.04±0.16^{a}	2.77±0.27^{ab}	2.73±0.14^{b}	2.78±0.16^{ab}
	Syneresis (%	2.99±0.16	2.85±0.27	3.02±0.14	2.91±0.16
	T2 (1.2% cinnamon)	3.05±0.16	2.64±0.27	3.06±0.14	3.20±0.16

Table 2. Antioxidant activity (Mean±SE) in yoghurt (percentage)

Sample	1st day	3rd day	5th day	7th day	Overall mean
Control	28.12±2.16^{Aa}	21.77±0.88 ^{Ab}	20.55±0.83 ^{Ab}	19.17±1.03^{Ab}	22.40±0.82^{A}
T1(0.6% cinnamon)	44.87±2.16^{Ba}	31.65±0.88^{Bb}	28.82±0.83^{Bc}	22.50±1.03^{Bd}	31.96±0.82^{B}
T2 (1.2% cinnamon)	54.77±2.16^{Ca}	34.50±0.88^{Cb}	30.53±0.83^{Bc}	23.60±1.03^{Bd}	35.85±0.82^{C}
Overall mean	42.59±2.16^{a}	29.30±0.88^{b}	26.63±0.83^{c}	21.76±1.03^{d}	

At 5 per cent levelmeans with same superscripts (small letters a-d within a row, capital letters A-C within a column) do not differ significantly.

Table.3 Sensory evaluation of (Mean ±SE) yoghurt samples

Sample	Appearance and colour	Body and texture	Flavour	Overall acceptance
Control	5.00±0.00	5.00±0.00	9.14±0.26A	19.14±0.26A
T1(0.6% cinnamon)	5.00±0.00	5.00±0.00	9.86±0.14B	19.86±0.14B
T2 (1.2% cinnamon)	5.00±0.00	5.00±0.00	9.43±0.30A	19.43±0.30A

At 5 per cent level means with same superscripts (capital letters A-B within a column) or without any superscripts do not differ significantly.

Fig. 1: Yoghurt Samples

Fig. 1a: 0.6% Cinnamon extract incorporated yoghurt

Fig. 1b: 1.2% Cinnamon extract incorporated yoghurt

Fig. 1c: Control yoghurt

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