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Effect of carrot juice and stabilizer on the physicochemical and microbiological properties of yoghurt

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A B S T R A C T

Yoghurt fortification with fruits and vegetables has high potential to improve the nutrients and health promoting effects of the yoghurt. The effects of carrot juice (0, 10, 15 and 20%) and gelatin stabilizer (0.5, 0.6 and 0.7%) (w/w, base milk) addition on the properties of 12 yoghurt samples in a 3 × 4 factorial arrangement were investigated. Addition of carrot juice increased pH and syneresis significantly, but decreased titratable acidity (TA) and total viable counts (TVC). The TA and TVC were higher than minimum recommended of 0.6% lactic acid and 6 $\log_{10}$ CFU g^{-1}, respectively for yoghurt. Coliform, yeast and mold counts were <10 CFU g^{-1}. Syneresis decreased with stabilizer addition ($p < 0.01$). With 10 to 20 percent carrot juice addition, the total carotenoid content (mg/kg) increased (6.73 and 10.26, respectively) compared to control (3.05) ($p < 0.05$). However, the effects of carrot juice and stabilizer additions on total phenolic contents and antioxidant ferric reducing power were insignificant ($p > 0.05$). The results showed that yoghurt with suppressed syneresis and improved nutritional and total carotenoids contents can be processed from 10 to 15 percent carrot juice and 0.7 percent stabilizer additions.

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1. Introduction

The development of diversified dairy products such as fruits and vegetables fortified yoghurt has a significant contribution for the dairy sector development in Ethiopia. The healthy food image of yoghurt is due to its probiotic effects which include protection against gastrointestinal upsets, enhanced digestion of lactose, decreased risk of cancer, lower blood cholesterol, improved immune response, enhanced short chain fatty acids (SCFAs) production, assimilation of protein and calcium (Granato, Branco, Cruz, Faria, & Shah, 2010; Gahruie, Eskandaria, Mesbahi, & Hanifpour, 2015).

Yoghurt with no added flavor is predominantly sour due to the lactic acid produced by fermentation. For better acceptance, fruits, flavoring agents and sweeteners are added to yoghurt to improve flavor balance and mask partially acetaldehyde flavor of yoghurt (Routray & Mishra, 2011). Various evidences demonstrate that fruits and vegetables intakes are associated with an improved health because of various nutrients and bioactive phytochemicals (Sun-Waterhouse, 2011). Thus, yoghurt are fortified with various fruits (Ayar, Sert, Kalyoncu, & Yazici, 2006; Oliveira et al., 2015), fruit seed extracts (Chouchouli et al., 2013) and vegetables (Puvanenthiran, Stevovitch-Rykner, McCann, & Day, 2014; Gahruie et al., 2015) to enhance positive health promoting effects of the yoghurt.

Carrot is rich in β-carotene and bears ascorbic acid, tocopherol and is classified as vitaminized food (Sharma, Karki, Singh, & Attri, 2012). It also bears carbohydrates, calcium, phosphorus, iron, potassium, magnesium, copper, manganese, sulfur and phenolic compounds, but it is deficient in protein and fat. Yoghurt is rich in protein, fat, calcium, potassium, B vitamins (B1, B2, B6, nicotinic and pantothenic acids) but is deficient in iron, vitamin C, carotenes and dietary fibers (Gahruie et al., 2015). Thus, combination of carrot juice and yoghurt will improve nutritional and functional food characteristics of the yoghurt. Salwa, Galal, and Neimat (2004) have studied the effect of carrot juice blending ratio on the shelf life and

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sensory properties of yoghurt and reported that shelf life and consumer acceptance were improved with 15% carrot juice addition. However, an attempt to reduce syneresis was not made which is a defect in yoghurt processing. To suppress syneresis, addition of stabilizers like gelatin or other hydrocolloids that function as a gelling agent or thickener provide good stability and desirable yoghurt textures (Routray & Mishra, 2011). Ares et al. (2007) showed addition of gelatin at a level of 0.6% (w/w) into yoghurt has suppressed incidence of syneresis. Therefore, in this work, the effects of carrot juice and stabilizer levels on the physicochemical, microbiological and functional (probiotic, total carotenoid, total phenolic and antioxidant ferric reducing power) properties of twelve yoghurt formulations are reported.

2. Materials and methods

2.1. Milk samples and ingredients

Fresh whole cow's milk was collected from Haramaya University dairy farm, Ethiopia. Fresh carrots (*Daucus carota* L. cv. Nantes) were purchased from farmer's field located near Haramaya University, stabilizer (gelatin with 240 Blooms manufactured in Brazil by Bake Mate) and sweetener (cane sugar) were purchased from a supermarket. Freeze-dried yoghurt starter culture (YC-X11 CHR HANSEN) was purchased from chemical suppliers (Nile Star Import and Export, Addis Ababa).

2.2. Experimental design

The experiment was conducted in triplicate in a completely randomized design of 3 × 4 factorial combinations of gelatin (0.5, 0.6 and 0.7 g per 100 g of milk) and carrot juice (0, 10, 15 and 20 g per 100 g of milk).

2.3. Preparation of carrot juice

Carrot roots were washed thoroughly, ends removed, peeled by sharp knife, cut longitudinally into halves and blanched at 90 °C for 5 min to tenderize carrot tissues and inactivate pectinase and peroxidase enzymes (Salwa et al., 2004). Carrot juice was extracted in a mechanical blender with sieves (Type 6001×, model No. 31JE35 6 × .00777, USA) and analyzed for moisture, total soluble solids (TSS), titratable acidity, total sugar, total phenolics and total carotenoid contents.

2.4. Yoghurt processing

Prior to yoghurt processing, all equipment used was sterilized in an autoclave after thorough wash cleaning. Heat sensitive materials such as plastic equipment were placed in boiling water for 30 min to kill vegetative cells on the material surface.

Starter preparation: The yoghurt starter culture was inoculated into fresh milk that was heated at 90 °C for 30 min. The inoculate was incubated at 45 °C until pH 4.6 was attained, stored overnight (4 °C) and then was used in the yoghurt processing.

Twelve yoghurt formulations were processed as described by Ayar et al. (2006). Dry ingredients (gelatin and 4% cane sugar on milk weight basis) were weighed and separately blended into three different gelatin stabilizer levels (0.5, 0.6 and 0.7%, w/w). Each dry ingredient blend was mixed thoroughly with fresh milk, filtered through cheese cloth and preheated to 50 °C to facilitate melting of gelatin and uniform mixing. The resulting premix was heat treated for 30 min at 85 °C and cooled to 45 °C in a 4 °C water bath. Then each of the three premixes was further divided into four equal portions to which carrot juice (0, 10, 15 and 20%, w/w on milk basis) was added. Maintaining a temperature of 43 °C, the resulting yoghurt samples were filled into coded screw capped glass jars, inoculated with 3% (w/w) yoghurt starter culture and incubated (Gallenkamp Incubator Plus Series, England) at 43 °C for a period of 2.5 h. To determine the incubation period and to monitor fermentation progress, a separate control yoghurt (yoghurt without carrot juice and 0.6% gelatin) was prepared to which pH probe was inserted, concurrently incubated in a thermostatically controlled water bath (43 °C). The incubation of the experimental yoghurt was terminated when the pH of the control yoghurt reached 4.7. The yoghurt samples were then immediately cooled by transferring them from the incubator to a refrigerator (4 °C) and then stored for 24 h. The effects of carrot juice and stabilizer addition levels on the physicochemical and microbiological characteristics were reported based on yoghurt samples analyzed in triplicate after 24 h of storage.

2.5. Proximate composition of milk and yoghurt

The total solids content of milk was determined according Richardson (1985) and that of yoghurt according to IDF (1991). The moisture content of both milk and yoghurt samples were determined by difference (AOAC, 1998 Method No. 990.20):

$$\%\text{Moisture} = 100 - \text{Total solids (TS\%)}$$

The fat contents of milk and yoghurt were determined using AOAC (1998) Method No 905.02 and solids-not-fat (SNF) by subtracting percentage of fat from total solids (TS %). Total protein (% N × 6.38) and ash contents were determined as described in AOAC (1998) Methods No. 991.20 and No. 945.46, respectively.

2.6. pH and titratable acidity (TA) of milk and yoghurt

The TA of both milk and yoghurt samples were determined by titrating with 0.1 N NaOH (Richardson, 1985) and TA was expressed as percentage of lactic acid (LA). The pH was determined by a glass electrode attached to the pH meter (model number 510 Cyber Scan, Eutech Instruments) with a temperature probe after calibrating (buffer solutions pH 4 and 7).

2.7. Chemical analysis of carrot juice

The total solids content of carrot juice was determined by oven drying (AOAC, 1998 Method 920.151), total soluble solids (TSS) by hand refractometer (Atago N1, USA) (AOAC, 1998 Method 932.12) and total sugar by colorimetric method (Somogyi, 1945). The TA was expressed as percentage of citric acid equivalent (AOAC, 1998 Method No 942.15), pH by a glass electrode attached to the pH meter (model pH 510, Oakton instruments USA).

2.8. Total phenolics content (TP) from carrot juice and yoghurt

2.8.1. From carrot juice

The TP content was determined by the Folin-Ciocalteu method after extraction from carrot juice with 30 mL of solvent (80% aqueous ethanol, containing 1% conc. HCl) in a conical flask, agitating in an orbital shaker (200 rpm, at 50 °C, for 2 h), filtering (Whatman No. 4) as described in Lima et al. (2005). The filtered extract was used for the determination of TP content and ferric reducing power.

The extract (100 μL) was mixed with 750 μL of Folin-Ciocalteu reagent, allowed to stand at room temperature for 5 min and mixed gently with 750 μL of 6% (w/v) sodium carbonate. A blank was made by mixing distilled water and reagents. After allowing to

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