



## Physicochemical and sensory characteristics of orange juice supplemented yogurt

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### ABSTRACT

In recent years, the development of enriched dairy products with fruits or fruit parts has been growing due to their potential health benefits and consumer's preferences. In this research, orange juice incorporated yogurt was elaborated, and the effect of orange juice incorporation was evaluated in terms of physicochemical and sensory attributes. Fresh orange juice was extracted using an electric juicer. Skim milk powder, starter culture, and sugar were used to prepare four yogurt samples, S1, S2, S3, and S4, containing 0%, 3%, 5%, and 7% orange juice, respectively. Syneresis, water holding capacity (WHC), pH, viscosity, and firmness of the samples were compared, and the sensory quality of the prepared yogurt was evaluated. With the increasing orange juice percentage, the syneresis of the yogurt increased. Sample S4 (7% orange juice supplemented yogurt) exhibited a higher syneresis value ( $11.37\% \pm 0.81$ ) than the other samples. Meanwhile, the WHC, pH, and viscosity decreased when a higher proportion of juice was assimilated to the yogurt. The lowest values for WHC, pH, and viscosity were possessed by S4 (7% orange juice), where the values were 53.20, 3.68, and 123.33 m Pas, respectively. The firmness of yogurt improved with the addition of higher orange juice content. In the sensory test, orange juice yogurt obtained higher scores than the control one. The panelists preferred S2 (3% orange juice) which got the highest scores for color, flavor, mouthfeel, taste, and overall acceptability among all the samples. The result exhibited an innovative consumer-based fruit yogurt with changes in its properties.

**Keywords:** Fruit yogurt, orange juice supplementation, syneresis, sensory attributes, water holding capacity



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## 1 Introduction

Nowadays, consumer's preference for nutritionally modified low-fat foods with functionality is growing (Bimbo et al., 2017). Consumers prefer dairy products the most as a functional food (Bimbo et al., 2017;

Ozer and Kirmachi, 2010). Different properties of milk products are extensively explored because dairy foods are the ideal medium for functional additives and ingredients (Hati et al., 2019; Beltrán-Barrientos et al., 2016). However, several health issues associated with dairy products such as lactose intolerance,

allergenicity, hypercholesterolemia have been reported (Munekata et al., 2020; Szilagyi and Ishayek, 2018). Nevertheless, yogurt is one of the most consumed dairy products in more than a century, with abundant health benefits (Aryana and Olson, 2017). It has been a part of the human diet for its nutritional profile and the presence of probiotic microorganisms (Pan et al., 2019). Probiotics are living microorganisms and, when consumed, provides several health benefits (Hill et al., 2014), including prevention and treatment of gastrointestinal issues (Dolin, 2009), improving the intestinal microbiota (Sun et al., 2011), developing immune system and lactose tolerance (Kim-mel et al., 2010). Due to its nutrient profile, yogurt is considered a nutrient-dense food and is a rich source of calcium that provides significant amounts of calcium in bio-available form. In addition, it includes milk proteins with a higher biological value and provides almost all the essential amino acids necessary to maintain good health. Consumption of yogurt also promotes bone health, reduces chronic diseases, and supports a healthy life cycle by improving the diet quality (Shiby and Mishra, 2013).

Yogurt is also valued for its organoleptic properties due to lactic acid fermentation (Cais-Sokolińska and Walkowiak-Tomczak, 2021). Routray and Mishra (2011) mentioned that the popularity of yogurt as a food component is linked to its sensory attributes. Considering the health benefits and consumer's perception, several researchers investigated the preparation of yogurt and often supplemented with fruits, fruit pulps, fruit seed extracts, and vegetables (Wallace and Giusti, 2008; Chouchouli et al., 2013; Perina et al., 2015; Costa et al., 2015; Dimitrellou et al., 2020). Adding these ingredients enhances flavor, includes bioactive phytochemicals, enzymes, and antioxidants, alters the microbiological, functional, and sensory properties, and improves health-promoting profile.

The incorporation of fruits endorses a healthy image of yogurt and has significantly contributed to yogurt consumption among all ages (Chandan and Shahani, 2006). Fruit juices are healthy, refreshing, and well-accepted by young and old consumers. Orange juice is popular because of its aroma and taste. Sweet orange (*Citrus sinensis* L. Osbeck) is a member of the Rutaceae family and a major source of vitamins, especially vitamin C, sufficient amount of folacin, calcium, potassium, thiamine, niacin, and magnesium. It is low in calories and full of nutrients; promotes clear, healthy skin as part of an overall healthy and varied diet. Usually, oranges are eaten fresh or used for making jam, jelly, juice, resins, and orange seed oil. Phytochemicals are present in orange and, when eaten favorably, modulate human metabolism, preventing chronic and degenerative diseases (Tripoli et al., 2007). A single orange is said to have about 170 phytonutrients and over 60 flavonoids with anti-tumor, anti-inflammatory, blood clot inhibiting, and

antioxidant properties, which help to benefit overall health (Cha et al., 2001). Thus, the addition of orange juice may provide some functional benefits to yogurt for human health.

However, a limited number of studies have been conducted on orange juice supplemented yogurt (Isah, 2016; Hossain et al., 2012). Besides the health benefits, some physical, chemical, and textural properties play a vital role in consumer preferences (Pan et al., 2019). To the best of the author's knowledge, no studies have yet been done to explore the effect of orange juice on syneresis, water holding capacity, viscosity, and firmness of yogurt. Therefore, the present study was designed to develop yogurt using skim milk powder with various percentages of orange juices and evaluate the influence of orange juice on the physicochemical and sensory properties of the yogurt.

## 2 Materials and Methods

### 2.1 Materials

Fresh orange (*Citrus sinensis* L. Osbeck), skimmed milk powder (brand- Marks, Diet: non-fat milk powder), and sugar (brand- Fresh, refined sugar) were collected from the local market; starter culture (YC-087, Cher. Hansen, inc., Milwaukee, WI) was purchased from a local supplier of Dinajpur. All the involved materials and chemicals were of analytical grade and were used from the Food Engineering and Technology laboratory, Hajee Mohammad Danesh Science and Technology University, Dinajpur, Bangladesh.

### 2.2 Preparation of reconstituted skim milk

Reconstituted skim milk was prepared by dispersing 14 g of skim milk powder in 100 mL distilled water. The dispersion was agitated using a magnetic stirrer for 30 minutes. Then the milk was heated in a thermostatically controlled water bath at 85 °C for 20 minutes and cooled to room temperature (28 ± 2 °C).

### 2.3 Preparation of orange juice

Fresh oranges were washed with clean tap water to remove unwanted materials and dirt from the surface. The orange skin was separated by using stainless steel knife. Seeds were removed, and juice was extracted by using a juicer (Walton WBL-13MX35, Bangladesh) without adding excess water. Afterward, the juice was filtered by using a filter paper and placed in a plastic container and stored at freezing temperature (−20 °C) for further use during yogurt preparation.

## 2.4 Starter culture preparation

Yogurt was prepared using commercial starter culture (YC-087, Cher. Hansen, Inc., Milwaukee, WI), containing mainly *L. bulgaricus* and *S. thermophiles*. Starter culture was prepared by adding 0.1 g of freeze-dried culture to 100 mL of reconstituted skim milk and mixed adequately by using an electric stirrer. The starter culture was incubated at 37 °C for 10 hours and stored in the refrigerator (4 ± 2 °C).

## 2.5 Yogurt preparation

Sugar (6% w/v) was added to the reconstituted milk, and the milk was stirred for 30 minutes using an electric stirrer for proper mixing (Fig. 1). Then, it was heated at 80–85 °C for pasteurization and cooled to room temperature (28 ± 2 °C). After cooling, 0%, 3%, 5%, and 7% orange juice was incorporated into the milk to prepare four yogurt samples and mixed well (Table 1). The juice was added before incubation with the starter culture, as Guven and Karaca (2002) suggested. Afterward, milk was incubated, adding 2% (w/v) starter culture. Fermentation was carried out at 37 °C for 10 hours, and the prepared yogurt was stored at 4 °C for further analysis.

**Table 1.** Formulation of yogurt

Ingredients	S1	S2	S3	S4
Distill water (mL)	100	100	100	100
Skim milk powder (%)	14	14	14	14
Sugar (%)	6	6	6	6
Starter culture (%)	2	2	2	2
Orange juice (%)	0	3	5	7

S1=Control yogurt, S2=Yogurt with 3% orange juice, S3=Yogurt with 5% orange juice, S4=Yogurt with 7% orange juice

## 2.6 Analysis of yogurt

The prepared yogurt samples were analyzed for syneresis, water holding capacity, pH, firmness, texture, and viscosity.

### 2.6.1 Syneresis

Syneresis or whey separation is an important physical test of yogurt quality. Whey separation from yogurt samples was performed using a previously used method mentioned by Supavitpatana et al. (2008) with some modifications. A 10 g yogurt sample was spread in a thin layer to cover the Whatman No.1 filter paper. The yogurt was filtered for 15 minutes. Then, the filtrate was collected, and the weight was recorded. The percentage syneresis was calculated

using the following equation:

$$\text{Syneresis (\%)} = \frac{\text{Wt. of filtrate}}{\text{Total wt. of yogurt}} \times 100 \quad (1)$$

### 2.6.2 Water Holding Capacity (WHC)

A modified centrifugation method was utilized to measure the water holding capacity due to a high external force (Guzman-Gonzalez et al., 1999). For this test, falcon tubes containing yogurt at equal weight were used. The falcon tubes were placed in the centrifuge machine (General centrifuge, MF-300) and centrifuged at 1500 rpm for 15 minutes. The surface whey expelled from yogurt was gently poured off and quantified. The percent WHC was calculated by the following formula:

$$\text{WHC (\%)} = \frac{W_s - W_w}{W_s} \times 100 \quad (2)$$

where  $W_s$  = weight of sample and  $W_w$  = weight of whey.

### 2.6.3 pH

An electrode pH meter (HI 2211 pH/ORP meter) was used to measure the pH of the yogurt. The pH meter was calibrated routinely with fresh pH 4 and 7 standard buffer (Behrad et al., 2009). Then, the pH meter electrode was directly dipped into the yogurt sample, and the pH was recorded immediately after taking out the samples from the incubator.

### 2.6.4 Viscosity

The viscosity of the samples was determined by using a rotational viscometer (VR-3000, model-L) at room temperature (28 ± 2 °C) with a spindle rotation of 100 rpm. The readings were recorded at 15 seconds of the measurement. The measurements were taken three times for each yogurt sample, and the readings were recorded as centipoises (m Pas) (Djordjevic-Denin et al., 2002).

### 2.6.5 Firmness

The firmness of yogurts is crucial in establishing consumer preference. The essential textural characteristics of yogurt are firmness and the ability to retain water. The firmness of the yogurt sample was measured with an instrumental compression/penetration test using a digital fruit penetrometer (GY 4) equipped with an extrusion cell of 11 mm diameter. All yogurts were measured at freezing conditions. The determination was carried out in plastic containers. The depth of the yogurt was 25 mm. It was expressed as the maximum force (N) as the test cell penetrated to a depth of 10 mm into the sample described by Mohamed and Morris (1987).

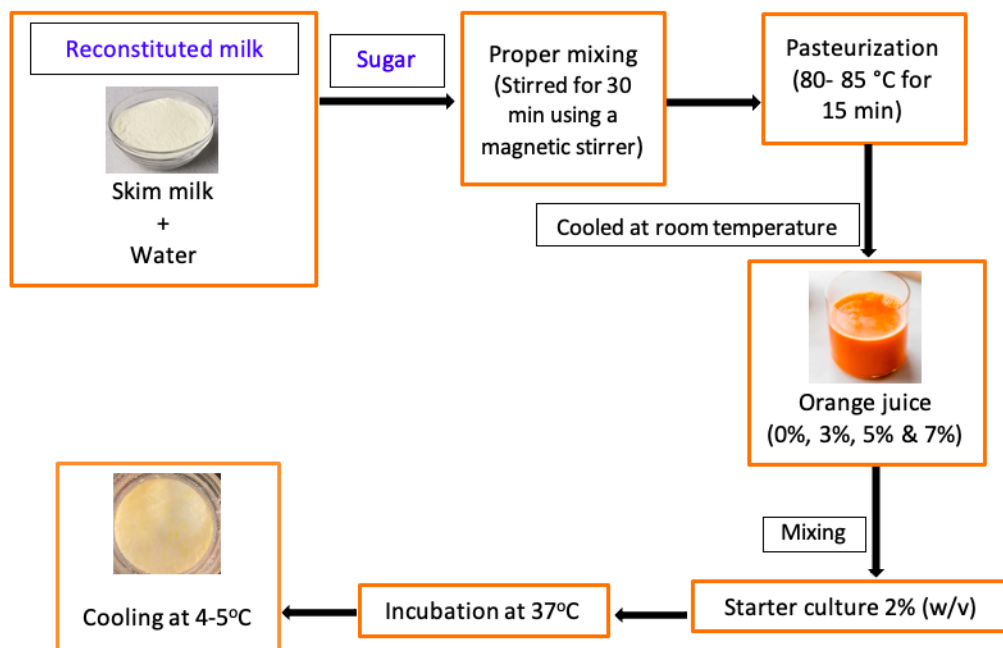


Figure 1. Flow diagram showing different steps of yogurt preparation

## 2.7 Sensory analysis of yogurt

A trained test panel of 30 members evaluated the consumer's acceptability of prepared yogurt of various combinations. Sensory characteristics of the product such as color, flavor, mouthfeel, taste, and overall acceptability were evaluated using a 1 to 9-point hedonic scale (9=like extremely, 8=like very much, 7=like moderately, 6= like slightly, 5= neither like nor dislike, 4= dislike slightly, 3= dislike moderately, 2=dislike very much, and 1= dislike extremely) (Amerine et al., 2013). Chilled samples of various combinations were presented to the panelists in identical plastic cups labeled in random order. The panelists recognized the yogurt only by code.

## 2.8 Statistical analysis

All the observation was replicated thrice. Results were prepared as means of  $\pm$  SEM. Comparisons of means were performed by one-way analysis of variance (ANOVA) followed by Duncan's test ( $P \leq 0.05$ ). Statistical analysis was run using the SPSS program (IBM Corporation, Inc. 2013, version 22).

## 3 Results and Discussion

### 3.1 Physiochemical properties

#### 3.1.1 Syneresis

Syneresis or whey separation is a common defect in evaluating yogurt quality. This defect affects the product shelf-life and the consumer's accept-

ability due to undesirable appearance (Dimitrellou et al., 2020). The syneresis values of orange juice incorporated yogurt are depicted in Fig. 2, and the lowercase letters indicate the values were significantly different ( $p \leq 0.05$ ). The syneresis gradually increased with more acidic juice incorporation ranging from 6.27% to 11.37%. Higher syneresis values were also found when white/red dragon fruit (Zainoldin and Baba, 2009) and carrot juice were added to yogurt (Kiros et al., 2016). Akyuz and Coskun (1995) mentioned that increasing syneresis is common in fruit yogurts. Several factors, including acidity, low fat, heat treatment, and incubation condition, can induce this increasing trend (Rani et al., 2012). Horne (1999) described that acidification of milk decreases the charges on caseins and weakens the electrostatic forces that hold micelles collected, thus cutting the steric stabilization provided by charged k-casein 'hairs' on the micelle surface. In the absence of steric stabilization, the attractive force between micelles becomes weak, which causes adhesiveness or stickiness and forms a weak particle gel. Researchers reported that rearrangement of the network of casein micelles was the leading cause of the yogurt syneresis (Anema et al., 2004; Vasbinder et al., 2003). Furthermore, the addition of fruit concentrate could decline the consistency of yogurt due to the reduction in the water-binding capacity of proteins (Ozturk and Oner, 1999). According to Penna et al. (2001), reduction in water holding capacity leads to more whey releases and increases the syneresis. However, increment of syneresis of yogurt is not desirable and needs to be overcome. Therefore, to achieve desired consistency and prevent syneresis, yogurt is often stabilized with



viscosity modifiers (such as starches, gums, gelatins, pectins) (Ramasmwamy and Basak, 1992; Amatayakul et al., 2006).

### 3.1.2 Water holding capacity (WHC)

Fig. 3 presented the WHC of the yogurt where the values are significantly different (the lowercase letters indicate the significant differences by Duncan's test at  $p \leq 0.05$ ). The WHC decreased with the addition as well as the increasing proportion of orange juice into the yogurt. The values were 72.03%, 60.92%, 58.62%, and 53.19% for S1, S2, S3, and S4 respectively. Usually, WHC and syneresis follow the opposite trend, and lower WHC contributes to higher syneresis, and this study experienced likewise. The lower WHC might be due to the higher acidity of orange juice, which could affect both soluble protein complex and the micelle bound (Xu et al., 2015). Ozturk and Oner (1999) reported fruit concentrate incorporation negatively influences the water-binding capacity of proteins. Similar results were found by other authors who concluded that the addition of concentrated fruit decreases the WHC and increases the whey separation of fruit-incorporated yogurt (Celik and Bakirci, 2003; Atasoy, 2009). Another researcher found that pH change during whey processing may alter the yogurt texture and the WHC (Sodini et al., 2006). However, the inclusion of stabilizers like gelatin can improve the WHC of yogurt (Amatayakul et al., 2006; Pang et al., 2015).

### 3.1.3 pH

pH indicates the presence of organic acid, and acidification is the primary mechanism for yogurt fermentation (Brabandere and Baerdemaeker, 1999; Zainoldin and Baba, 2009). The recommended range for the pH of yogurt is pH 4.6 or lower (Frye, 2013). Table 2 shows the pH variations before and after incubation of yogurt. The pH of prepared yogurt drastically decreased after incubation due to the orange juice incorporation, where there was no significant difference in the pH among the samples. However, the highest (3.77) and lowest (3.68) pH was found for S1 and S4, respectively. The results indicated that the yogurts become more acidic with the additional percentage of orange juice. A similar result was found when grape juice or dragon fruit was added to yogurt (Calvo et al., 2002; Zainoldin and Baba, 2009). The declining pH value could be explained as the proto cooperation of the bacterial strains (*L. bulgaricus* and *S. thermophiles*) (Brabandere and Baerdemaeker, 1999). The nutrient-enriched matrix and the optimum incubation environment support the rapid growth of the bacterial strains (Lourens-Hattingh and Viljoen, 2001). The high bacterial metabolic activity ferments lactose and produces lactic acid, acetaldehyde, diacetyl, and

formic acid, which accumulate and decrease the pH of yogurt (Gaspar et al., 2013).

### 3.1.4 Viscosity

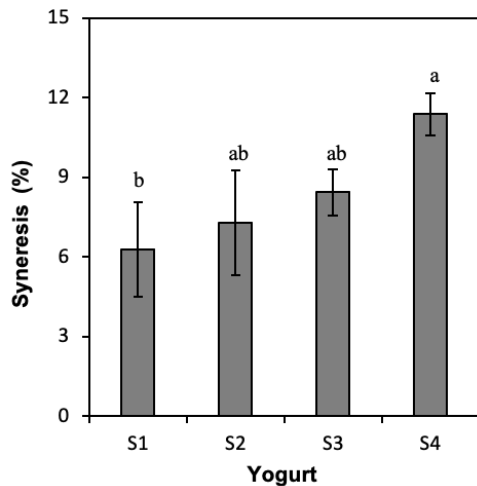
The viscosity of all the yogurt samples was statistically different ( $p \leq 0.05$ ). From Fig. 4, a negative trend can be seen where the viscosity of yogurt decreased with the increasing percentage of orange juice from 0% to 7% (declining range from 168.67 to 123.33 m Pas). The lowercase letters on the bars indicate the significant differences by Duncan's test at  $p \leq 0.05$ . S4 (7% orange juice) obtained the lowest, and S1 (control) exhibited the highest viscosity. Decreasing viscosity is common in fruit yogurts (Akyuz and Coskun, 1995). The water content in orange juice might have reduced the consistency of the yogurt; therefore, a lower viscosity was obtained with the additional orange juice. Researchers mentioned that the percent of dry matter present in the yogurt (Ayar and Gurlin, 2014) and the recovery of the starter (Lee and Lucey, 2010) could affect the viscosity. The processing steps of yogurt can also influence the viscosity, as explained by Keating and White (1990).

### 3.1.5 Firmness

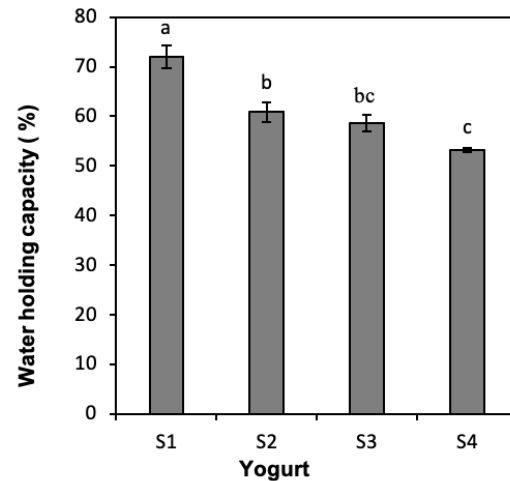
Firmness is considered a texture parameter determined by the arrangement and structure of the components; influenced by several factors, including composition and production processes; and an important indicator to evaluate the physical and sensory attributes of dairy products (Walia, 2013). As seen from Fig. 5, the firmness increased with the higher amount of orange juice inclusion (the values were 4.73, 4.83, 5.1, and 5.7 N for S1, S2, S3, and S4, respectively). The lowercase letters on the bars indicate the significant differences by Duncan's test at  $p \leq 0.05$ . This result might be due to the composition of orange juice, which consists of hydroxyl group and phenolic acid. The hydroxyl group has a strong affinity for caseins (Yuksel et al., 2010) in yogurt (whey protein). The phenolic group and protein interaction could create soluble complexes, which would affect the texture of coagulated yogurt (Kumar and Mishra, 2003). Researchers also reported the compositional influence of pomegranate juice powder on yogurt (Pan et al., 2019). However, some researchers claimed that other processing parameters such as temperature, fat content, homogenization condition, starter culture, and incubation could influence the firmness of yogurt (Lucey et al., 1997; Sodini et al., 2004).

## 3.2 Sensory evaluation

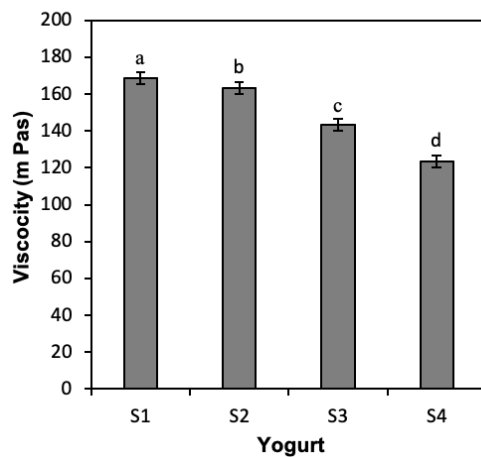
The prepared yogurt samples were subjected to sensory evaluation and compared at a 5% confidence level for color, flavor, mouthfeel, taste, and overall



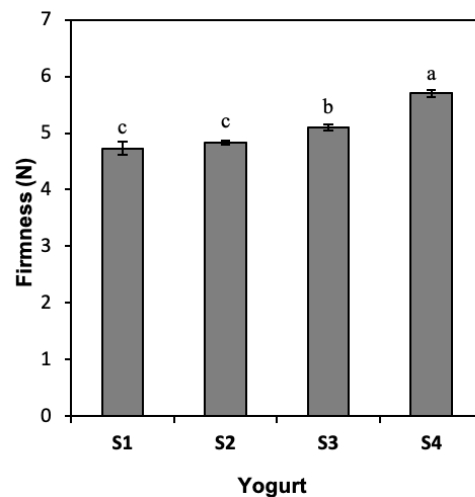
**Figure 2.** Syneresis percentage in yogurt with 0%, 3%, 5%, and 7% orange juice



**Figure 3.** Water holding capacity of yogurt with 0%, 3%, 5%, and 7% orange juice



**Figure 4.** Viscosity of yogurt with 0%, 3%, 5%, and 7% orange juice



**Figure 5.** Firmness of yogurt with 0%, 3%, 5%, and 7% orange juice

acceptability. Based on Table 3, the orange juice incorporated samples differed significantly from the yogurt without orange juice. The hedonic scores of S2, S3, and S4 were statistically similar for color, flavor, and taste preferences; nevertheless, they differed in mouth feel and overall acceptability. However, S2 (3% orange juice) exhibited the highest score for all the attributes. On the other hand, control sample S1 (0% orange juice) scored the lowest for all properties. Some researchers found the highest overall acceptability scores for a higher amount of fruit pulp incorporation into yogurt (Amal et al., 2016), whereas, Zahedi et al. (2015) added 7%, 8%, and 9% orange peel flavonoid in yogurt and found the maximum sensory scores for 8%. These results, as mentioned above, indicated that the component or fruit part added plays a vital role in the sensory attributes. In this work, the addition of orange in the form of juice could have attributed to the current finding. Moreover, the color and aroma of

orange and the texture of yogurt samples could have combinedly contributed to the obtained consumer's perceptions scores. Finally, the results outlined that S2, S3, and S4 were significantly more acceptable than S1; and all the panelists preferred S2 the most.

## 4 Conclusion

In this study, skim milk-based orange juice (at 0%, 3%, 5%, and 7%) supplemented yogurt was prepared. After incorporating orange juice, a significant change was found in the syneresis, WHC, viscosity, and firmness of the yogurt. All the parameters were influenced when more orange juice was added to the yogurt. The percent syneresis was increased, whereas the WHC and viscosity were decreased compared to the control yogurt (without orange juice). The firmness also improved with the addition of orange juice. Nonetheless, there was no significant differ-

**Table 2.** pH value of yogurt with 0%, 3%, 5%, and 7% orange juice

Sample	pH (before incubation)	pH (after incubation)
S1	6.51 ± 0.03a	3.77 ± 0.01a
S2	6.4 ± 0.03ab	3.73 ± 0.03a
S3	6.36 ± 0.05b	3.7 ± 0.01a
S4	6.29 ± 0.01b	3.68 ± 0.04a

S1=Control yogurt, S2=Yogurt with 3% orange juice, S3=Yogurt with 5% orange juice, S4=Yogurt with 7% orange juice; The lowercase letters indicate the significant differences by Duncan's test at  $p \leq 0.05$

**Table 3.** Sensory attributes of orange supplemented yogurt with 0%, 3%, 5%, and 7%

Samples	Color	Flavor	Mouthfeel	Taste	Overall acceptability
S1	6.30 ± 0.675b	5.90 ± 0.876b	5.90 ± 0.994b	6.00 ± 0.667b	6.20 ± 0.632c
S2	8.30 ± 0.823a	7.90 ± 0.876a	7.10 ± 0.876a	8.20 ± 0.919a	8.20 ± 0.632a
S3	8.20 ± 0.789a	7.40 ± 0.843a	6.50 ± 0.850ab	7.60 ± 2.066a	7.50 ± 0.850b
S4	8.00 ± 0.667a	7.10 ± 0.994a	5.90 ± 0.738b	7.20 ± 1.033a	7.00 ± 0.667b

All values are mean ± SD [Here, S1= Control Yogurt, S2= Yogurt with 3% orange juice, S3= Yogurt with 5% orange juice, S4=Yogurt with 7% orange juice] SCORE: Like extremely=9; Like very much=8; Like moderately=7; Like slightly=6; neither like nor dislike=5; Dislike slightly=4; Dislike moderately=3; Dislike very much=2; Dislike extremely=1]. The lowercase letters are indicating the significant differences by Duncan's test at  $p \leq 0.05$ .

ence in the pH of the samples. Furthermore, the sensory analysis revealed higher preferences for orange yogurt than the control yogurt, where S2 with 3% orange juice was the most preferred yogurt by the consumers. The results concluded that orange juice incorporation affected the yogurt properties and the consumer's perception of yogurt consumption. From this study, the potentiality of the commercial production of orange-flavored skim milk yogurt could be suggested. However, further shelf-life studies are required to evaluate the alterations in the properties of yogurt during storage. In addition, the interaction between probiotic cells and food components studies would also be advantageous to light up the potential health benefits of orange juice incorporated yogurt.

## Conflict of Interest

The authors declare that there is no conflict of interests regarding the publication of this paper.

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