

**J. Dairy Sci. 93:4471–4479 doi:10.3168/jds.2009-2582** © American Dairy Science Association<sup>®</sup>, 2010.

# Sensory and microbiological quality of yogurt drinks with prebiotics and probiotics

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# ABSTRACT

The popularity of dairy products fortified with prebiotics and probiotics continues to increase as consumers desire flavorful foods that will fulfill their health needs. Our objectives were to assess the sensory profile of drinkable yogurts made with prebiotics and probiotics and to determine the viability of the probiotics in the vogurt drink over the duration of storage. Thirteen trained descriptive panelists evaluated 10 yogurt drinks on a 16-point category scale. Three selected prebiotics, soluble corn fiber, polydextrose, and chicory inulin, were each present individually at an amount to claim an excellent source of fiber (5 g of fiber/serving) or a good source of fiber (2.5 g of fiber/serving) in 6 different yogurt drinks. Three additional vogurt drinks contained 5 g of each of the separate prebiotics along with a mixture of the selected probiotics (Bifidobacterium lactis Bb-12 and Lactobacillus acidophilus LA-5). A control sample with no prebiotics or probiotics was also included in the experimental design. Data were analyzed by ANOVA, Fisher's least significant difference, and principal component analysis. Survival of the probiotics in the yogurt drinks during a 30-d refrigerated storage period was also analyzed. Results showed that clover honey aroma, buttermilk aroma, butter aroma, sweetness, sourness, chalky mouthfeel, and viscosity were identified as significant attributes in the yogurt drinks. Total variance explained by the principal component analysis biplot of factors 1 and 2 was 65%, which showed yogurt drinks with soluble corn fiber and inulin varying by the sweet versus sour attributes and yogurt drinks with polydextrose varying by the mouthfeel attributes. The viability study determined a 2- to 3-log decrease in the survival of probiotics in all of the yogurt treatments during a 30-d refrigerated storage period. Based on the results of the current study, only the polydextrose treatment

would be an acceptable vehicle to deliver the probiotic health effects at the end of the 30-d storage period. **Key words:** sensory, yogurt, probiotic, prebiotic

## INTRODUCTION

Yogurt is among the most common dairy products consumed around the world, and its sensory attributes have a large effect on consumer acceptability (Saint-Eve et al., 2006). Drinkable yogurt, categorized as stirred yogurt with a low viscosity, is a growing area of interest based on its convenience, portability, and ability to deliver all of the health and nutritional benefits of stirred or set yogurt (Eder, 2003; Thompson et al., 2007). The low viscosity is obtained through high agitation, which breaks the coagulum after the fermentation period, before the product is bottled and refrigerated (Tamime and Robinson, 1985). The Food and Drug Administration (FDA, 2008) standard of identity for yogurt drinks specifies > 8.25% milk solids-not-fat and fat levels to satisfy nonfat yogurt (<0.5%), low-fat yogurt (2%), or yogurt (>3.25%) before the addition of other ingredients (Chandan et al., 2006). A typical low-fat yogurt drink available in the United States has 8.0 to 9.5%milk solids-not-fat and contains 5 to 12% added sugar. Yogurt drink pH varies from 4.0 to 4.5 (Tamime and Robinson, 1985; Chandan et al., 2006). In regard to flavor, strawberry is the most popular yogurt flavor followed by other fruit flavors (Thompson et al., 2007). Currently, few nonflavored vogurt drinks are available in the United States.

As the popularity of yogurt products continues to grow, manufacturers are continuously investigating value-added ingredients such as prebiotics and probiotics to entice health-conscious consumers. Probiotics are referred to as "live microorganisms, which when administered in adequate amounts confer a health benefit on the host" (FAO/WHO, 2001). Lactobacillus and Bifidobacteria species are the most common types of probiotics. Prebiotics are classified as "non-digestible food ingredients that beneficially affect the host by selectively stimulating the growth and/or activity of one

Received July 22, 2009.

Accepted June 7, 2010.

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or a limited number of bacteria in the colon, and thus improve host health" (Gibson and Roberfroid, 1995). Currently, the most widely accepted prebiotics include fructooligosaccharides and galactooligosaccharides (ISAPP, 2009). When prebiotics are combined with probiotics, their relationship is classified as synbiotic. This combination can improve the survival rate of the probiotics and provide additional health benefits to the host (Collins and Gibson, 1999).

Sensory analysis of yogurt drinks with prebiotics and probiotics is needed for manufacturers looking to incorporate the healthful ingredients into their products. Inclusion of probiotics has been shown to not significantly alter the sensory properties of dairy products (Hekmat and Reid, 2006); however, prebiotics such as inulin have the ability to be a fat substitute, bulking agent, low-calorie sweetener, and texture modifier when added to yogurt, therefore potentially altering the sensory perception of the product (Guggisberg et al., 2009). The growing number of possible prebiotics needs to be evaluated for their sensory effect before commercial inclusion. Previous studies determined that selected probiotics do not alter the sensory profile of prebiotic-containing yogurt (Hekmat and Reid, 2006; Kailasapathy, 2006). However, it is unknown if this is true for all probiotics and whether this applies to prebiotic-containing yogurt drinks.

The viability of the probiotic strains in the yogurt drink matrix is another area of interest when investigating the quality of yogurt drinks with novel prebiotics and probiotics. Currently, there is no standard of identity for probiotics (Sanders et al., 2007). The suitable level of viable probiotic cells remains obscure with no current regulatory requirements. Additionally, the minimum dose for a given health benefit likely varies for individual probiotics (Sanders et al., 2007). Regardless, it has been suggested that foods with probiotics should contain from  $10^6$  to  $10^8$  cells/g (not including starter culture) and remain at this level for the duration of the product's shelf life (Fonden et al., 2000). To effectively deliver the optimal level of bacterial cells to the consumer, it is critical that viable cell counts are assessed and appropriate measures are taken to ensure the survival of the bacteria.

The main objectives of this study were (1) to produce an appealing vanilla control yogurt drink similar to commercial products that will serve as a basis of comparison for yogurt drinks with added prebiotics and probiotic cultures, (2) to determine the effects of prebiotics and probiotics added to the model yogurt formulation on the flavor, aroma, and texture properties using descriptive analysis, and (3) to investigate the survival of the probiotic cultures as affected by the prebiotics tested in the yogurt drinks with prebiotics over a 30-d refrigerated storage period.

## MATERIALS AND METHODS

## **Development of Yogurt Drink**

For the yogurt drink process used in this study, pasteurized and homogenized skim milk (Schnucks Skim Milk, Schnucks Market Inc., St. Louis, MO) was used as the main dairy ingredient. During the first stage of formulation, sucrose (C+H Pure Cane Sugar Granulated White, C+H Sugar Company Inc., Crockett, CA) was added to the milk at a relatively low level of 5%(5-12% recommended) because inclusion of prebiotics has been shown to contribute to the sweetness of yogurts (Guggisberg et al., 2009). The milk and sugar were then heated and agitated (ThermoMix TM 21, Vorwerk USA Co., Altamonte Spring, FL) at 70°C for 20 min to mimic a pasteurization step. In the yogurt drinks with prebiotic inclusion at a low or high level, the prebiotic was incorporated after 10 min of heating to ensure full dispersion. This time and temperature combination was selected based on process stability data available for Litesse polydextrose (Litesse II Super Improved Polydextrose Fcc, Danisco USA Inc., Ardsley, NY; Beer et al., 1991), which allowed us to use a milder heat treatment than the typical heat treatment applied in yogurt manufacture of 85°C for 30 min. After the pasteurization step, the yogurt drink was cooled in the freezer  $(0-5^{\circ}C)$  until a temperature of  $42^{\circ}C$  was reached and the drink was then ready to be inoculated with starter cultures at a level of 0.02%. When the probiotics Bifidobacterium lactis Bb-12 and Lactobacillus acidophilus LA-5 (BB-12 and LA-5 Nutrish a/B probiotic mix, Chr. Hansen Inc., Milwaukee, WI) were included in the formulation, the level of starter cultures was reduced to 0.01%. Both starter cultures and probiotics were initially present as frozen pellets and were first thaved and then prepared as a 10% dilution in water before inoculation. The procedures for preparing and adding the probiotics were determined based on manufacturer recommendations. Probiotics were present as a 50:50 ratio mix of Bb-12 and LA-5 and were added at the same time of the starter cultures. The starter cultures were a mix of Streptococcus thermophilus and Lactobacillus delbrueckii ssp. bulgaricus (F-DVS YC-X11, Yo-Flex, Chr. Hansen Inc.). After inoculation, the yogurt drink was then agitated for 30 min to disperse the cultures while maintaining a temperature of 42°C. The product was then placed in an incubator (Isotemp Standard Incubator 637D, Fisher Scientific, Pittsburgh, PA) for fermentation at 42°C for around 5 to 6 h until pH 4.3

#### YOGURT QUALITY WITH PREBIOTICS AND PROBIOTICS

		Without probiotics					With probiotics $(Pro)^1$			
		Polydextrose (PDX, 90% fiber)		Soluble corn fiber (SCF, 66% fiber)		Chicory inulin (89% fiber)		PDX + Pro	$\begin{array}{c} \mathrm{SCF} \\ + \mathrm{Pro} \end{array}$	Inulin + Pro
Item	Control	Low	High	Low	High	Low	High	High	High	High
Prebiotics <sup>2</sup> (%)	0	1.23	2.45	1.70	3.34	1.24	2.48	2.45	3.34	2.48

Table 1. Experimental design of yogurt sensory study for 10 total treatments

<sup>1</sup>Probiotics were Lactobacillus acidophilus LA-5 and Bifidobacterium lactis Bb-12 (Chr. Hansen, Milwaukee, WI).

<sup>2</sup>Amounts of prebiotics used were dependent on the percentage of fiber in the prebiotic to allow for the claims of a good source of fiber (2.5 g/ serving) or an excellent source of fiber (5 g/serving; FDA, 2008) for a 240-mL serving size.

to 4.4 was obtained. The pH was initially measured at 5 h of fermentation and every 20 min thereafter until the desired pH was reached. Fermentation time varied within yogurt drink replicates, suggesting that prebiotic or probiotic addition was not a significant factor for determining fermentation time. After fermentation, the coagulum was broken through an additional agitation step at 42°C for 30 min. During this time, powdered natural and artificial vanilla flavor (Natural & Artificial Vanilla Flavor Powder, lot# S090833, Flavors of North America-FONA, Geneva, IL) was added to the yogurt drink at a level of 0.20% to provide an acceptable consumer product for consumption. After the final agitation step, the yogurt was poured into sanitized Mason jars (1 L, Hearthmark Inc., Muncie, IN) and refrigerated for at least 24 h before serving. To determine specific amounts of the ingredients, a serving size of 240 mL was assumed for the product.

## Experimental Design

Ten vogurt drink treatments were included in the study design. Table 1 depicts the different yogurt drinks by percentage of prebiotic included in the formulation. Three prebiotics were used in this study, polydextrose (Litesse II Super Improved Polydextrose FCC, Danisco USA Inc., Ardslev, NY), chicory inulin (Beneo GR, Orafti Active Food Ingredients, Tienen, Belgium), and soluble corn fiber (Promitor Soluble Corn Fiber, Tate & Lyle, Decatur, IL). Each prebiotic was present at a low level and a high level in the yogurt drink based on the amount of fiber it contained. The low level corresponded to the amount required to claim the product as a good source of fiber (2.5 g/serving) and the high level to that required to claim the product as an excellent source of fiber (5 g/serving) as determined by the US Food and Drug Administration (FDA, 2008). Three of the 10 vogurt drinks contained the high level of the prebiotics along with a mixture of the selected probiotics, 0.02% of *Bifidobacterium lactis* Bb-12 and *Lacto*bacillus acidophilus LA-5. A control yogurt drink was also present in the design; it contained no prebiotics or probiotics. A yogurt drink containing only probiotics was not included in the study because numerous studies have shown no sensory effect of probiotic inclusion alone in yogurt (Atunes et al., 2005; Hekmat and Reid, 2006).

# Subjects for Descriptive Panel

Thirteen panelists (4 male, 9 female, aged 21–29 yr) participated on the panel. Subjects were initially recruited based on interest, availability, nonsmoking status, and lack of food allergies.

Following recruitment, subjects were further screened with a basic tastes test where they were asked to identify the taste associated with several solutions at low concentrations. Solutions were served in 29.6-mL plastic cups with lids (Solo Cup Company, Urbana, IL). Sucrose, citric acid, sodium chloride, and caffeine were added at a low level to water to be identified as sweet (0.70% sucrose solution), sour (0.05% citric acid solution), salty (0.10% NaCl solution), or bitter (0.02% caffeine solution) by the panelists (Mojet et al., 2001). Six solutions were presented, with citric acid being presented twice and spring water once as a blank.

The prospective panelists were further screened by 6-n-propyl-2-thiouracil (**PROP**)-impregnated filter paper prepared according to Zhao et al. (2003) and asked if they were able to perceive a bitter taste when placing the paper on their tongue. The PROP paper was served in 29.6-mL plastic cups with lids (Solo Cup Company). Subjects who identified 3 out of the 6 basic taste solutions correctly and who perceived the bitter taste associated with the PROP test were further asked to participate on the panel.

# Panel Training

The 13 panelists selected for the descriptive panel participated in twenty 1-h training sessions. The first 5 sessions focused on term generation and introduction to references. During the following weeks, the panel refined and defined the attributes they generated and practiced scaling references in relation to the intensity of the attribute within the yogurt products they tasted. The 10 yogurt drinks in the experimental design (Table 1) were presented and evaluated an equal number of times throughout the training process. Panelists also developed a rinsing protocol to eliminate any carryover effect while evaluating the yogurt. This consisted of carbonated water (Schweppes Tonic Water, Plano, TX) followed by cool spring water (Absopure, Plymouth, MI). During the 15 d of training, the panel came up with 12 attributes that most consistently described the vogurt products (Table 2) and practiced evaluating the products in a roundtable setting to facilitate discussion. Subsequently, they moved on to individual booth training for 4 d.

## Sample Evaluation

Panelists completed two 1-h final evaluations on separate days, in which they evaluated each yogurt drink product presented in the design (Table 1) in duplicate. The panelists sat around a table and refamiliarized themselves with the references and the reference intensities (Table 2). The panelists then went into individual booths equipped with Compusense Five data acquisition system (Version 4.8, Compusense, Guelph, Ontario, Canada) and evaluated the 10 yogurt samples. Each sample was served monadically to keep the temperature constant  $(0-5^{\circ}C)$ . All samples were given to panelists in 59.1-mL cups (Solo Cup Company) labeled with a random 3-digit code, and products were presented in a randomized order across panelists. Panelists were instructed to rinse before tasting each sample. To minimize fatigue, there was a 10-min break in between sample 5 and 6 during each evaluation hour. Evaluation was conducted under incandescent lighting and at room temperature ( $\sim 24^{\circ}$ C). Each attribute was evaluated on a 16-point category scale that ranged from 0 to 15.

# Assessing Viability of Commercial Probiotic Cultures in Yogurt Drink

Five yogurt drinks were tested for viable cell counts of *B. lactis* Bb-12 and *Lb. acidophilus* LA-5. The yogurt drinks included in the viability study were a control (no pre- or probiotics), control with only probiotics, polydextrose (5 g of fiber) with probiotics, soluble corn fiber (5 g of fiber) with probiotics, and inulin (5 g of fiber) with probiotics. Yogurt drinks were stored in a refrigerator at 4°C. The 5 yogurt samples were plated on d 1, 10, 20, and 30 in triplicate for each probiotic. The method used for selective enumeration of viable *L. acidophilus* LA-5 and B. lactis Bb-12 cells was obtained from Chr. Hansen (P-10 and P-12 technical bulletin). To determine the survival of the Lb. acidophilus culture, 0.5 mL of sterilized 0.02% clindamycin stock solution (Clindamycin HCl, Biomol Research Labs Inc., Plymouth Meeting, PA) was added per liter of deMan, Rogosa, and Sharpe (MRS) agar (Lactobacilli MRS Broth, Hardy Diagnostics, Santa Maria, CA; and agar granulated, Fisher Scientific, Fair Lawn, NJ). Bifidobacteria were selectively grown on MRS agar that contained 5 mL of 0.01% dicloxacillin stock solution (Dicloxacillin sodium salt, MP Biomedicals LLC, Solon, OH), 10 mL of 11% lithium chloride stock solution (Acros Organics, Morris Plains, NJ), and 5 mL of 10% cysteine hydrochloride stock solution (L-cysteine hydrochloride hydrate, Acros Organics) per liter of medium. The spiral plate (Eddy Jet-Spiral Plater, IUL Instruments, Barcelona, Spain) technique was used for enumeration of both probiotics. All plates were incubated anaerobically  $(90\% N_2, 5\%)$  $H_2$ , and 5%  $CO_2$ ) at 37°C for 3 d before colonies were enumerated.

#### Statistical Analyses

The data were analyzed using ANOVA by the GLM procedure and the mean separation test by the Fisher's least significant difference (LSD) with a 95% confidence level (SAS version 9.1, SAS Institute Inc., Cary, NC). Principal component analysis (**PCA**) biplots were constructed using XLSTAT version 2008 (Addinsoft USA, New York, NY).

## **RESULTS AND DISCUSSION**

## **Descriptive Analysis**

A total of 7 out of the 12 attributes generated by the panel were found to be significantly different (P < 0.05) across the yogurt drinks. These 7 attributes included clover honey aroma, buttermilk aroma, butter aroma, sweetness, sourness, chalky mouthfeel, and viscous mouthfeel (Table 3). Panelists were a significant source of variation (P < 0.05) for all attributes. This could be caused by scaling differences among the panelists, which is typically seen in descriptive analysis panels. Replication by sample and panelist by sample interactions were not significant sources of variation for any of the attributes, indicating that the panelists were consistent in their evaluations, and the panel as a whole agreed.

Specific product differences from the descriptive panel can be identified in Table 4. For chalky mouthfeel, only one of the prebiotic beverages (inulin low) had a significantly greater chalky mouthfeel compared with the

Modality	Term	Definition	Reference preparation	Reference product	$\begin{array}{c} \text{Reference} \\ \text{intensity}^1 \end{array}$
Aroma	Vanilla	The aroma of diluted vanilla extract	1 g of vanilla extract in 300 mL of water [5 g into 147.87-mL (5-oz.) cup]	Durkee Pure Vanilla Extract (ACH Food Companies Inc., Memphis, TN)	10
	Clover honey	The sweet aroma of diluted clover honey	0.85 g of honey in 30 mL of water [place in 147.87-mL (5-oz.) cup]	Great Lakes Clover Honey (Great Lakes, Onsted, MI)	11
	Buttermilk	The sour aroma of diluted buttermilk	0.88 g of buttermilk in 10 mL water [place in 147.87-mL (5-oz.) cup]	Prairie Farms 1% Buttermilk (Prairie Farms, Carlinville, IL)	10
	Butter	The aroma of solid butter	0.3 g of solid butter [place in 147.87-mL (5-oz.) cup]	Schnucks Butter (Schnucks Market Inc., St. Louis, MO)	11
Aroma-by-mouth	Vanilla	The aroma of diluted vanilla flavoring while in mouth	1.5 g of vanilla powder in 250 mL of water [20 mL served in 29.57-mL (1-oz.) cup]	Natural and Artificial Vanilla Flavor Powder, Lot# S090833 (Fona, Geneva, IL)	10
Taste and aftertaste	Sweet	The taste of sucrose in solution while sample is in mouth	6.3 g of sugar in 600 mL of water [20 mL served in $29.57$ -mL (1-oz.) cup]	C+H Pure Cane Sugar Granulated White (C+H Sugar Company Inc., Crockett, CA)	9
	Sour	The taste of citric acid in solution while sample is in mouth	0.35 g of citric acid in 500 mL of water [20 mL served in 29.57-mL (1-oz.) cup]	Anhydrous granular citric acid (Tate & Lyle, Decatur, IL)	11
	Sour aftertaste	The immediate aftertaste of lactic acid solution after the sample is expectorated	2 g of lactic acid in 800 mL of water [20 mL served in 29.57-mL (1-oz.) cup]	88% Lactic Acid (LD Carlson Company, Kent, OH)	11
	Cheese aftertaste	The instantaneous aftertaste of ricotta cheese after the sample is expectorated	1 tsp. of part-skim ricotta cheese served in 29.57-mL (1-oz.) cup	Schnucks Ricotta Part-Skim Cheese (Schnucks Market Inc.)	10
Mouthfeel	Chalky	The mouthfeel of crushed calcium carbonate tablets dispersed in skim milk	25 calcium carbonate tablets ground in 250 mL of skim milk [20 mL served in 29.57-mL (1-oz.) cup]	TopCare Antacid Calcium Chewable Tablets, Extra Strength, Assorted Fruit (Topco Associates LLC, Skokie, IL)	10
	Viscous	The instantaneous thickness of heavy cream in mouth	Heavy cream [20 mL served in 29.57-mL (1-oz.) cup]	Schnucks Heavy Whipping Cream (Schnucks Market Inc.)	10
Afterfeel	Dryness	The sensation felt on the tongue and sides of the mouth after the sample has been expectorated	1.0 g of tannic acid in 1,000 mL of water	Tannic Acid (Sigma Chemical, St. Louis, MO)	11

# Table 2. Descriptive terms with definitions and references with reference preparation and intensity

 $^{1}$ References were rated iteratively over 5 d to generate a reference intensity and averaged over those 5 d of evaluation to give a mean value for the reference on a 16-point category scale that ranged from 0 to 15.

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		Replication (R)	Panelist (P)		Interaction		
Modality	Attribute			Sample (S)	$\mathbf{R} \times \mathbf{P}$	$\mathbf{R} \times \mathbf{S}$	$\mathbf{P}\times\mathbf{S}$
Aroma	Vanilla	1.94	5.02***	1.05	1.43	0.77	0.94
	Clover honey	1.28	9.06***	$2.69^{**}$	1.25	0.72	0.85
	Buttermilk	1.32	8.70***	$3.64^{***}$	1.27	0.54	1.02
	Butter	1.90	$15.99^{***}$	$1.97^{*}$	1.77	0.56	1.06
Aroma-by-mouth	Vanilla	1.77	$13.33^{***}$	1.73	0.93	0.40	1.09
Taste and aftertaste	Sweet	$6.48^{*}$	$12.69^{***}$	$2.10^{*}$	1.76	1.00	0.92
	Sour	4.09	$10.77^{***}$	$2.13^{*}$	$2.82^{**}$	1.28	1.02
	Sour aftertaste	0.02	9.24***	1.51	$1.96^{*}$	1.15	1.31
	Cheese aftertaste	0.77	$26.16^{***}$	0.82	$2.98^{**}$	2.01	1.09
Mouthfeel	Chalky	$5.22^{*}$	$5.68^{***}$	$2.10^{*}$	1.56	0.65	0.65
	Viscous	2.57	11.64***	$3.91^{***}$	$4.21^{***}$	0.71	0.96
Afterfeel	Dryness	0.02	$19.41^{***}$	1.00	0.95	1.90	0.98

Table 3. Analysis of variance	(F-values)	) of 12 sensory attributes rated across 10 yogurt drinks

\*P < 0.05; \*\*P < 0.01; \*\*\*P < 0.001.

control that contained no prebiotics or probiotics (P < 0.05). Beverages containing polydextrose at a high level and inulin at high and low levels were significantly more viscous than the control (P < 0.05). Increasing the concentration of prebiotics, especially inulin, has been shown to increase the viscosity and other mouthfeel characteristics of products (Guggisberg et al., 2009). Inulin yogurt drinks exhibited the highest sweet taste perception compared with the control (P < 0.05). Inulin, in particular short-chain inulin, has been shown to have a sweetness profile similar to that of saccharose and can significantly increase the sweetness level of a product (Villegas et al., 2010).

Probiotics have been shown in previous studies to not alter the sensory properties of yogurt (Atunes et al., 2005). However, as illustrated in Table 4, the addition of probiotics into the yogurt drinks that contained prebiotics at a high level did cause sensory attribute differences. In particular, when adding probiotics to the polydextrose and soluble corn fiber yogurt drinks, the chalkiness of the products became significantly greater than that of the control beverage (P < 0.05). Other specific product differences were found when adding probiotics. The polydextrose with probiotics yogurt drink had the lowest sweet clover honey intensity compared with the control (P < 0.05), and also had the highest sour buttermilk aroma compared with the other probiotic and prebiotic yogurt drinks (P < 0.05). The effect of polydextrose as a sweetener and its effect on increasing sweet aroma have been shown to be dependent on the product. In an antioxidant extract study, its effect at increasing sweetness was almost imperceptible compared with other sweeteners used in the study (Ares et al., 2009). The butter aroma intensity of the yogurt drink with soluble corn fiber at the high level became significantly lower (P < 0.05) when probiotics were incorporated in the beverage. Similarly, the addition of the same probiotics, Lb. acidophilus LA-5 and *B. lactis* Bb-12, to drinkable yogurts made with goat milk reduced the intensity of several attributes

Table 4. Mean intensity ratings for significant attributes

$Sample^1$		Aroma attribute		Taste at	tribute	Mouthfeel attribute	
	Clover honey	Buttermilk	Butter	Sweet	Sour	Chalky	Viscous
Control	$8.38^{\mathrm{ab}}_{\mathrm{ba}}$	$9.12^{\mathrm{ab}}_{\mathrm{ba}}$	7.92 <sup>ab</sup>	8.23 <sup>d</sup>	$8.58^{\mathrm{a}}_{\mathrm{ab}}$	$6.81^{\mathrm{d}}$	$7.62^{\mathrm{de}}_{\mathrm{bal}}$
PDX High	$8.08^{\mathrm{bc}}$	$8.65^{\mathrm{bc}}$	$7.54^{\mathrm{abcd}}$	8.81 <sup>abcd</sup>	$8.19^{\mathrm{ab}}$	$7.08^{\rm cd}$	$8.31^{\rm bc}$
PDX Low	$7.92^{\rm bc}_{\rm p}$	$8.77^{\mathrm{bc}}$	$7.15^{cd}$	$8.54^{\text{bcd}}$	$8.19^{\mathrm{ab}}$	$7.15^{\text{bcd}}$	$8.04^{\text{bcde}}$
Inulin High	$7.88^{ m bc}$	$8.77^{ m bc}$	$7.65^{\mathrm{abcd}}$	$9.27^{ m ab}$	$7.88^{ m ab}$	$7.27^{ m abcd}$	$8.69^{\mathrm{ab}}$
Inulin Low	$9.15^{\mathrm{a}}$	$7.73^{ m d}$	$7.77^{ m abc}$	$9.35^{\mathrm{a}}$	$8.12^{\mathrm{ab}}$	$7.77^{ m ab}$	$9.00^{\mathrm{a}}$
SCF High	$7.69^{ m bc}$	$8.35^{ m bc}$	$8.11^{a}$	$8.38^{ m cd}$	$8.42^{\mathrm{a}}$	$7.04^{\rm cd}$	$7.96^{ m cde}$
SCF Low	$8.23^{ m bc}$	$8.31^{\rm cd}$	$7.46^{\text{abcd}}$	$9.27^{\mathrm{ab}}$	$7.50^{\mathrm{b}}$	$7.08^{\rm cd}$	$7.42^{ m e}$
PDX + Pro	$7.5^{\circ}$	$9.73^{\mathrm{a}}$	$7.77^{\rm abc}$	$9.00^{ m abcd}$	$8.54^{\mathrm{a}}$	$7.88^{\mathrm{a}}$	$8.42^{\rm abc}$
Inulin + Pro	$7.88^{ m bc}$	$8.85^{ m bc}$	$7.38^{ m bcd}$	$9.31^{\mathrm{ab}}$	$8.08^{\mathrm{ab}}$	$7.42^{\mathrm{abcd}}$	$7.85^{ m cde}$
SCF + Pro	$8.27^{ m bc}$	$8.23^{ m cd}$	$7.04^{\mathrm{d}}$	$9.15^{ m abc}$	$7.58^{\mathrm{b}}$	$7.65^{ m abc}$	$8.23^{ m bcd}$

<sup>a-e</sup>Means in the same column with the same letter are not significantly different (P < 0.05).

 $^{1}$ PDX = polydextrose; SCF = soluble corn fiber; Pro = probiotics; high and low refer to levels of prebiotics added.

#### YOGURT QUALITY WITH PREBIOTICS AND PROBIOTICS

		$\mathrm{Treatment}^2$								
C.	Control + Pro		PDX + Pro		SCF + Pro		Inulin + Pro			
Storage time (d)	LA-5	Bb-12	LA-5	Bb-12	LA-5	Bb-12	LA-5	Bb-12		
1 10 20 30	$\begin{array}{c} 1.70 \times 10^8 \\ 4.87 \times 10^7 \\ 4.87 \times 10^6 \\ 4.80 \times 10^5 \end{array}$	$\begin{array}{c} 1.03 \times 10^8 \\ 4.0 \times 10^7 \\ 3.15 \times 10^6 \\ 6.00 \times 10^5 \end{array}$	$\begin{array}{c} 1.08 \times 10^8 \\ 3.33 \times 10^7 \\ 3.90 \times 10^6 \\ 1.00 \times 10^6 \end{array}$	$\begin{array}{c} 1.21 \times 10^8 \\ 4.43 \times 10^7 \\ 3.15 \times 10^6 \\ 1.10 \times 10^6 \end{array}$	$\begin{array}{c} 2.03 \times 10^8 \\ 3.83 \times 10^7 \\ 6.51 \times 10^6 \\ 6.30 \times 10^5 \end{array}$	$\begin{array}{c} 1.06 \times 10^8 \\ 3.47 \times 10^7 \\ 7.30 \times 10^6 \\ 4.40 \times 10^5 \end{array}$	$\begin{array}{c} 2.76 \times 10^8 \\ 1.80 \times 10^8 \\ 6.02 \times 10^6 \\ 2.87 \times 10^5 \end{array}$	$\begin{array}{c} 2.07 \times 10^8 \\ 1.47 \times 10^8 \\ 8.60 \times 10^6 \\ 1.87 \times 10^5 \end{array}$		

Table 5. Survival (cfu/mL) of Bifidobacterium lactis Bb-12 and Lactobacillus acidophilus LA-5 in yogurt drinks during 30-d storage<sup>1</sup>

<sup>1</sup>Control treatment with no prebiotics/probiotics had zero growth for all 30 d.

 $^{2}$ Pro = probiotics; PDX = polydextrose; SCF = soluble corn fiber.

(Uysal-Pala et al., 2006). Causes for the differences in the sensory attributes in prebiotic yogurt drinks with probiotics are undetermined. However, it is known that probiotics have been shown to increase organic acid concentrations (lactic and acetic acid) and proteolysis during fermentation and subsequent storage of set yogurts (Donkor et al., 2007). The addition of prebiotics to a probiotic-containing yogurt also contributes to alterations in organic acid concentrations (lactic and acetic acids) and proteolysis during fermentation and subsequent storage (Donkor et al., 2007; Vasiljevic et al., 2007). Additionally, the effects on organic acid

Biplot (axes F1 and F2: 64.56 %)

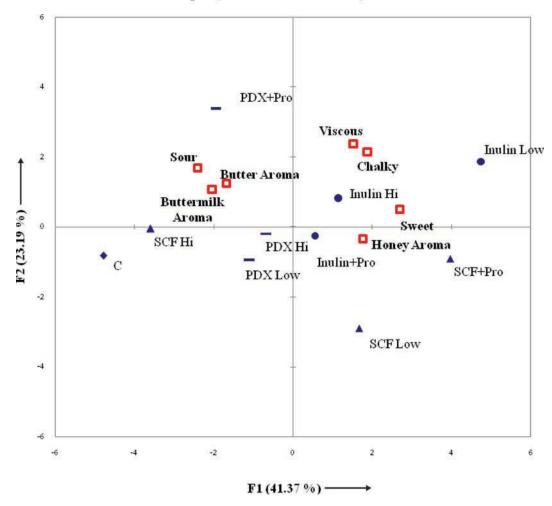


Figure 1. Principal component analysis biplot by the covariance matrix of the mean sensory ratings.  $\Box$  = attribute;  $\blacktriangle$  (SCF) = soluble corn fiber; - (PDX) = polydextrose;  $\bullet$  = inulin; and  $\bullet$  (C) = control; Pro = probiotics; Hi and Low = high and low levels of prebiotics added, respectively. Color version available in the online PDF.

production and proteolysis vary based on the specific prebiotic and probiotic added (Donkor et al., 2007; Vasiljevic et al., 2007).

The PCA biplot of the yogurt drinks (Figure 1) illustrates specific differences among the products. The biplot explains 64.6% of the total variation, with factor 1 on the x-axis explaining 41.4% of the data and factor 2 on the y-axis explaining 23.2%. Factor 1 was mostly accounted for by sweet and honey aroma versus sour, buttermilk aroma, and butter aroma based on the high loadings of these attributes on this factor. Factor 2 was characterized by the significant mouthfeel attributes, viscous and chalky. The yogurts with soluble corn fiber and inulin varied predominantly by factor 1; as we changed the amounts of prebiotics or added probiotics to these yogurt drinks, the sweetness versus sourcess taste and aroma and butter aroma attributes were affected the most. If we altered the amount of polydextrose in the product, the sensory attributes most affected were the mouthfeel attributes, in particular, chalkiness. Previous studies have shown the capability of polydextrose to provide an increase in viscosity and mouthfeel perception in various foods (Mitchell, 2004). Solutions of polydextrose have been shown to have a higher viscosity than other sweetener solutions, which enables the prebiotic to provide desirable mouthfeel qualities important when reducing sugar and fats (Mitchell, 1996).

## Viability of Probiotic Cultures in Yogurt Drink

Table 5 shows the survival of the probiotic strains *B. lactis* Bb-12 and *Lb. acidophilus* LA-5 during a 30-d refrigerated storage period. The control treatment with no prebiotics or probiotics demonstrated zero growth throughout the testing. There was an approximately 2-to 3-log loss in the total number of viable cells for both probiotics in all of the yogurt treatments during the 30 d. Several yogurt studies have determined similar results, showing that the survival of probiotic bacteria is often low in yogurt (Dave and Shah, 1997; Kailasapathy, 2006). The low pH of yogurt and postacidification of yogurt, further reducing the pH, contribute to the low viability of probiotics (Kailasapathy, 2006).

All yogurt treatments maintained between  $1.03 \times 10^8$ and  $2.76 \times 10^8$  cfu/mL of each probiotic after 1 d of refrigerated storage. By d 10, all treatments decreased by approximately 70%, except for the inulin treatment, which maintained a higher survival rate. Twenty days of refrigerated storage resulted in a decrease of approximately 85% of viable cells from d 10 in all of the yogurt treatments for both probiotics. At the end of the refrigerated storage period (d 30), *B. lactis* Bb-12 viable counts were  $1.10 \times 10^6$ ,  $4.40 \times 10^5$ ,  $1.87 \times 10^5$ , and  $6.00 \times 10^5$  cfu/mL for probiotic-containing yogurt with polydextrose, soluble corn fiber, inulin, and no prebiotic control, respectively. Lactobacillus acidophilus LA-5 viable counts were  $1.00 \times 10^{6}$ ,  $6.30 \times 10^{5}$ ,  $2.87 \times 10^{5}$ , and  $4.80 \times 10^5$  cfu/mL for probiotic-containing yogurt with polydextrose, soluble corn fiber, inulin, and no prebiotic control, respectively. All yogurt drink treatments lost more than 99% of viable bacteria during the 30 d. Variable results have been found when incorporating inulin in probiotic yogurt. Growth of L. acidophilus and L. rhamnosus have been shown to be enhanced with inulin (Sadek et al., 2004); however, similar to the results in this study, *Lb. acidophilus* LA-5 has been shown to not be stimulated by inulin in acidophilus-bifidus yogurts (Ozer et al., 2005). The minimal suggested level of viable probiotics at the time of consumption is approximately 10<sup>6</sup> cfu/mL of product (Adhikari et al., 2003). Therefore, based on the results of the current study, only the polydextrose treatment would be an acceptable vehicle to deliver probiotic health effects at the end of the 30-d yogurt drink storage.

Several methods to increase the survival of probiotics in yogurt have been recommended. Some of these include microencapsulation (Kailasapathy, 2006), a 2-step fermentation process with the probiotic fermentation first (Shah, 2000), and addition of ingredients known to increase viability such as cysteine, whey powder, and casein hydrolysates (Dave and Shah, 1998; Gomes et al., 1998; Adhikari et al., 2003). However, addition of such survival-enhancing ingredients may also affect the sensory characteristics of the yogurt, especially the texture (Kailasapathy, 2006). No specific viability differences were seen between the 2 probiotics in this study.

### CONCLUSIONS

This study demonstrated the effect of adding healthpromoting ingredients into a yogurt drink system. The prebiotics inulin, soluble corn fiber, and polydextrose were shown to alter the sensory properties of the yogurt drink when incorporated at different levels. When probiotics were incorporated, additional sensory changes were identified. This study can be used as a reference for yogurt manufacturers looking to incorporate novel prebiotics and probiotics in their products for the sensory effects of the ingredients at relevant levels. The viability study revealed a 2- to 3-log decrease in survival of the probiotics, with or without prebiotics, after 30 d of refrigerated storage. It is recommended that additional methods or ingredients be used to ensure the viability of probiotics in the product because several studies now prove their low survival rate. Future research could include microencapsulating the probiotics to determine if their survival rate is increased in the novel prebiotic yogurt drink and to determine if the sensory characteristics of the drink become affected.

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