ABSTRACT

NAKAMURA, RYAN PERRY. The Role of Sugar Reduction on Flavor and Acceptance of School Lunch Chocolate Milk. (Under the direction of Dr. MaryAnne Drake).

Our objective was to investigate sucrose replacement in school lunch chocolate milk with nutritive and nonnutritive sweeteners with and without lactose hydrolysis of milk on adult and child acceptance. In experiment 1, chocolate milk was formulated with nutritive and nonnutritive sweeteners (allulose, mixed sugar syrup, stevia, monk fruit, or sucralose) to the iso-sweet intensity of a current school lunch chocolate milk formulation with 3.69% sucrose (w/v). In experiment 2, lactose-hydrolyzed chocolate milk was formulated with nutritive and nonnutritive sweeteners (allulose, mixed sugar syrup, stevia, monk fruit) to the iso-sweet intensity of 3.69% sucrose (w/v). In experiment 3, lactose-hydrolyzed chocolate milk was formulated with sweetener blends of allulose, monk fruit and stevia to the iso-sweet intensity 3.69% sucrose (w/v). Iso-sweetness for all experiments was determined using magnitude estimation scaling and confirmed by 2-alternative forced choice paired comparison tests. Sensory properties of chocolate milks in each experiment were documented by descriptive analysis (DA) and temporal dominance of sensations (TDS). Consumer acceptance of chocolate milks was evaluated by young adult consumers for all experiments, and by child consumers for experiment 3. Sweeteners in unhydrolyzed chocolate milk (experiment 1) were differentiated ($p<0.05$) from the sucrose control chocolate milk due to bitter and metallic tastes inherent in the sweeteners. Liking scores for chocolate milks sweetened with allulose, mixed sugar syrup and sucralose were not different ($p>0.05$) from the sucrose control chocolate milk while liking scores for chocolate milks sweetened with stevia and monkfruit were lower than the sucrose control ($p<0.05$). The application of lactose-hydrolysis to milk (experiment 2) increased sweetness and reduced the amounts of each sweetener needed for isosweet taste by approximately one-third compared to

unhydrolyzed chocolate milk. Consumer acceptance of lactose hydrolyzed chocolate milks were not different ($p>0.05$). The application of sweetener blends in lactose hydrolyzed chocolate milk resulted in minimal differences by DA and TDS, and no differences ($p>0.05$) in overall liking to the sucrose control chocolate milk among adult and child consumers. The application of blends of nonnutritive sweeteners in combination with lactose hydrolysis proved effective at mitigation of sensory differences from sucrose sweetened chocolate milk while providing a lactose free no added sugar chocolate milk. The findings from this study can be useful for milk manufacturers in accomplishing the objective of a school lunch milk that is appealing to parents and children.

The Role of Sugar Reduction on Flavor and Acceptance of School Lunch Chocolate Milk

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DEDICATION

I dedicate this paper to my family who pushed me to be the best I can be and have supported me throughout my life. To Mom and Dad, thank you for everything, I am so grateful to have you two as my role models in life, encouraging me to chase my dreams while reminding myself to enjoy the moments as well. Andrew, thank you for always supporting me and providing me with guidance as I have grown into my own self. Your pursuit of a higher education pushed me to want to stand equal to you and pursue my own masters, as well as compete with you once more. You all have been my motivation to do my best at NC State and I love you all for helping me complete this goal.

BIOGRAPHY

Ryan Perry Nakamura was born in Berkeley California to his parents Lee and Catherine. Growing up in Oakland, CA with his older brother Andrew, Ryan enjoyed his time playing sports and working in his father's Japanese fish market where he found his passion for food science. After graduating from high school, Ryan pursued his B.S. degree in Food Science at the California Polytechnic State University of San Luis Obispo (SLO). During his time at Cal Poly SLO, Ryan became very involved in the department, participating in the Cal Poly Food Science Production Team and the Mars Product Development Competition Team. After graduating, Ryan decided to further his education with a Masters in Food Science in Dr. MaryAnne Drake's lab. With his Master's degree, Ryan hopes to make a positive impact on the food industry.

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Introduction

Milk consumption by children and adolescents has decreased since the 1970s based on the proportion of children consuming milk, the number of servings consumed per day, and the portion size of each serving of milk (Dror and Allen, 2014). The decline in milk consumption among children was not compensated with an increase in consumption of other dairy products, which contain essential nutrients important for the growth and development of children and adolescents (Baker et al., 1980; Cavadini et al., 2000; Black et al., 2002). Children who avoid dairy products are more likely to have inadequate calcium intake and poor bone health, which could impact adult health status and susceptibility to health issues (Huth et al., 2006; Weaver, 2010). Flavored milk, like chocolate and strawberry milk, is offered in schools to increase dairy consumption with children, which has a similar nutrient profile to plain milk but contains added sugar that contributes unwanted calories to school diets. Studies have addressed the reduction of the added sugar content of school lunch chocolate milk by sugar replacement with nonnutritive sweeteners and lactose hydrolysis (Li et al., 2014, 2015a; 2015b; Sipple et al., 2020). There has been comparatively less research conducted on the sensory analysis of flavored milk sweetened with nutritive and nonnutritive sweeteners, but numerous studies on the health impact of flavored milk on children and adults. This review will examine previous research on flavored school lunch milk as well as existing research on milk to explore how analytical and sensory methods can be used to increase consumer acceptance of school lunch milk.

Child Nutrition with Milk

Adequate nutrition during early stages of life is important for the growth and development of children, and milk and dairy products provide 51% of the dietary calcium intake for children of the ages 1.5 to 4.5 y; an age range where bone mineral content and bone mineral

density increase rapidly and peak in early adulthood (Wells, 2000). Calcium consumption via intake of dairy rich foods like yogurt, milk and cheese is important because humans continue to grow even after childhood and into early adolescence, and continued dairy consumption can lead to increases in bone mineral density, bone mineral content, and bone size (Chan et al., 1995; Bonjour et al., 1997; Huth et al., 2006). For children two years and older, milk is the number one food contributor for calcium, potassium, phosphorus, and vitamin D (Wells, 2000; Murphy et al., 2008; Rafferty and Heaney, 2008). Children who consume cow's milk are more likely to have a higher dietary calcium intake, larger skeletal structure, and higher bone mineralization (bone mineral content and bone mineral density) compared to children who did not consume cow milk (Baker et al., 1980; Black et al., 2002; Xueqin et al., 2004; Berkey et al., 2005). Bone growth and development during childhood has an important role in reducing the risk of fracture and susceptibility to osteoporosis during adulthood (Sandler et al., 1985; Kalkwarf et al., 2003; Golden et al., 2014). The 2010 US Dietary Guidelines for Americans recommended that U.S. children and young adults consume 3 cups of milk or milk products to meet the recommended calcium (1,000 mg for 4 to 8 year old, 1,300 mg for 9 to 13 year olds) (Institute of Medicine (US) Committee to Review Dietary Reference Intakes for Vitamin D and Calcium, 2011). Fulgoni et al. (2011) reported that to replace the calcium content from 1 serving of milk products (302.3 mg calcium per cup), it would require 1.1 servings of fortified soy beverage, 1.2 servings of bony fish, or 2.2 servings of leafy greens. The removal of one serving of dairy products slightly lowered the protein (7g), total fat (5.2g) and saturated fat (3.1g) intake, but significantly lowered the calcium (-229 mg/d) , and vitamin D (-1.5 μg/d) and phosphorus intake (Fulgoni et al., 2011). The consumption of milk and dairy products is important for the growth and development of children.

Beverage consumption provides important energy and nutrients in children's diets, and is responsible for 1 out of every 7 calories consumed by Americans ages 2 to 19 y (Berkey et al., 2004). For children ages 2 to 11 y, milk is the predominant source of beverage calories, but the majority of young adults ages 12 to 19 y change to sweetened beverages as the predominant source of beverage calories, with milk representing 22% of total beverage consumption among U.S. youth, followed by soft drinks (20%) and 100% juice (7%) (Dietary Guidelines Advisory Committee, 2020). Harnack et al. (1999) reported children and adolescents who were high consumers of sugar-sweetened soft drinks had lower intake of calcium, riboflavin, vitamin A and C, and phosphorus and folate. The consumption of soft drinks increased energy intake levels, which can contribute to obesity, and displace milk in the diets of children and adolescents. The majority of the U.S. population far exceeds the recommended limits of percent energy intake from added sugar, causing the higher likelihood of health related issues like obesity, hypertension, and cardiovascular diseases (Ludwig et al., 2001; Reid et al., 2007; de Ruyter et al., 2012).

Flavored Milk Nutrition

Flavored milk contains the same essential nutrients as plain milk: protein, calcium, potassium, phosphorus, vitamin A and B12, iodine and riboflavin, and children are more likely to consume milk when flavored milk is made available over plain milk (Murphy et al., 2008; Fayet-Moore, 2016). Fayet-Moore (2016) reported that children who consumed flavored milk had a similar micronutrient intake to children who consumed plain milk, and a higher nutrient intake compared to milk nondrinkers (Murphy et al., 2008). Peckham et al. (2021) found that students who selected fat-free chocolate milk consumed 8.5% more of their milk compared to students who selected low-fat white milk, resulting in 1.5 more grams of protein and 17 more milligrams

consumed. Johnson et al. (2002) reported that flavored milk consumption was positively associated with calcium, phosphorus and fiber intake, but negatively associated with vitamin A and C, and folate intake. Vanselow et al. (2009) found no significant changes to the BMI of chocolate milk consumers over a 5-year period but did report a significant inverse association between BMI and white milk consumption.

Flavored milk is a healthier alternative to sugar-sweetened beverages like soda and juice because it has less sugar, while providing a nutritional profile of protein and micronutrients identical to plain milk (Fayet-Moore, 2016). Johnson et al. (2002) reported that children who consumed flavored milk had higher calcium intakes with similar energy intake from total fat and added sugar intake compared to children who were non-consumers of flavored milk. Results from this study were possibly due to the result of lower intakes of soft drinks and fruit drinks by the children who consumed flavored milk. Murphy et al. (2008) reported similar added sugar intake between flavored milk drinkers and milk nondrinkers among children and adolescents aged 2 to 18 years. In Frary et al. (2004), the consumption of presweetened cereals and sweetened dairy products, like flavored milk, increased mean added sugar intake for children aged 6 to 11 years, but showed no association among adolescents. Sweetened dairy products like flavored milk can increase the number of dairy servings and the calcium intake among children, and increase the likelihood to meet the recommended adequate intake of calcium (Frary et al., 2004).

Child Perception of Milk

Studies have been published regarding child perception of plain and flavored fluid milk. Henry et al. (2015) found that children value health, environment, (physical and social), convenience routine/habit and variety/choice when asked about factors that influenced their milk

intake. In a focus group study of school-aged children, Henry et al. (2015) noted that the concept of taste was highlighted as important for consumption of milk, which is consistent with De Pelsmaeker et al. (2013) who reported that children prefer and consume more flavored milk than plain milk due to taste preference. De Pelsmaeker et al. (2013) also reported that children valued taste and health as important food choice motives, but significantly prioritized taste over health in terms of food consumption, with only 20% of children preferring plain milk to flavored milk. Thompson et al. (2007) also found that most Caucasian and Hispanic participants preferred flavored milk over plain milk due to flavor.

In addition to taste, the physical and social environment are important factors in children's ability to access and consume chocolate milk at home and at school. Henry et al. (2015) reported that physical environment factors consisted of the physical location of flavored milk being made available, for example, convenience stores were located near schools so students could purchase milk on their own. Social environmental factors were determined by relative or familial influence on the availability and acceptance of milk for the children. De Pelsmaeker et al. (2013) reported that parents provided plain milk for 40% of the children, and flavored milk for 17% of the children, despite children more likely to consume and prefer flavored milk over plain milk. The convenience of milk for children is very important as the easier it is to access milk, the higher likelihood that children will consume it. Children identified milk as a staple beverage in most households and perceived it as a routine but expressed a need for variety/choice in their beverages (Henry et al., 2015). Henry et al. (2015) also reported that convenience made a significant difference in child consumption of milk. It was noted that children preferred to not spend their own money on milk at school, especially when white milk was available free at home. Sipple et al. (2021) found that children prefer a specific milk due to

familiarity (e.g. if their parents had previously purchased it) or the appearance of the package (package type and graphics) via focus groups with school-aged children. Some children noted that milk from cartons "tasted funny" due to the paper packaging and had negative associations with the milk brand or type of package (Sipple et al., 2021). The size of the container and the number of servings was noted as the driver for selection among children and correlated to the amount of milk children perceived they could consume (Sipple et al., 2021). Focus groups also revealed that the presence of vitamins, calcium, increased protein content, percent fat labeling, and organic labeling positively influenced the health perceptions among the children, while higher levels of sugar and calories detracted from the health perceptions (Sipple et al., 2021).

Studies have reported that children understand the nutritional consequences of consuming flavored milk that contains sugar. Thompson et al. (2007) reported that 38% of Hispanic children in the study would limit their chocolate milk consumption because it was unhealthy to consume too much sugar. Children perceived milk as a healthy beverage while understanding that too much fat and sugar is not healthy for them as well (Henry et al., 2015). The preference of milk in children varies based on the influence from their parents and their own self-choice, and the decision to ban chocolate milk from school lunches needs to be carefully considered and weighed against the total dietary impact.

Velázquez et al. (2021) evaluated the hedonic sensitivity of children to dairy products (vanilla milk, chocolate-flavored milk, and vanilla yogurt) with reduced sucrose content. Velázquez et al. (2021) found that sucrose reduction up to 27% in chocolate flavored milk and yogurt and up to 19% in vanilla yogurt did not cause significant changes to the children's hedonic liking. Li et al. (2015a) found that 43.4% sugar reduction using monk fruit or monk fruit in chocolate milk were acceptable among child and adult milk consumers.

Published studies have explained how nutrition labels and packaging play an important role on the consumption and acceptance of dairy foods among children and adults. Ares et al. (2022) found that packaging claims largely influenced the preferences and food choices of parents and children and are necessary to protect children's health. Yoo et al. (2017) found that sugar claims significantly increased children and adolescents expected liking score, suggesting the consumers have a positive attitude towards sucrose reduction in dairy products. Ares et al. (2010) investigated the influence of non-sensory attributes on consumer's choice of functional over regular yogurts. This study found that brand, price and health claims had a significant impact on consumer choice and perception (Ares et al., 2010). Ares et al. (2016) used rating-based and choice-based conjoint with children to determine the influence of label design on school-aged children perception of snack foods like yogurt.

Parent Acceptance of Flavored Milk

Parents' perception and attitudes towards flavored milk and their drivers of decision-making are critical because they are responsible for purchasing and serving beverages to their children, along with allowing their children to partake in school lunch meal programs. When purchasing chocolate milk for their children, Li et al. (2014) reported that parents are most concerned with the type of sweetener and fat content of the flavored milk over brands, additional claims, organic/non-organic, or sugar content. Using a conjoint analysis, Li et al. (2014) reported that parents preferred chocolate milk with reduced sugar to no sugar added and regular sugar content, and preferred natural nonnutritive sweeteners for sugar reduction. Three different segments of parents were identified in the conjoint analysis for chocolate milk consumers (Li et al., 2014). Segment 1 was identified as the "Traditional parents" who preferred chocolate milk sweetened with sugar at the regular sugar content and with all natural/conventional milk, without

the use of non-caloric/nonnutritive or artificial sweeteners. Segment 2 consumers were the "all natural and organic parents" who preferred reduced sugar chocolate milk with natural noncaloric/nonnutritive sweeteners. Segment 3 consumers were the "health-conscious parents" who preferred all-natural, low-fat, or fat-free chocolate milk with reduced or no added sugar. Segment 3 consumers preferred low-fat and skim milk sweetened with natural, noncaloric sweeteners. Li et al. (2014) suggested that parents believe dairy products were essential for families due to the variety of nutrients in milk and milk was the major dairy product in the household.

Kim et al. (2013) confirmed that consumer acceptance of chocolate milk with adults was influenced by brand and fat content, with fat content being the major influencer for purchase of chocolate milk, followed by brand and sugar content. When participants actually tasted the milk, intrinsic factors like taste, sugar and fat content became most important when choosing their chocolate milk choice instead of extrinsic factors like brand name and packaging labels. Kim et al. (2013) also reported that adults preferred chocolate milk with 1 or 2% fat content for purchase but preferred the flavor of whole fat chocolate milk when tasting. This is consistent with Childs and Drake (2009) who evaluated Cheddar cheese preferences and Li et al. (2014) on chocolate milk where U.S. adults preferred low fat milk or cheese over full-fat or fat-free products conceptually, suggesting that U.S. consumers are interested in healthy food choices, but are not willing to sacrifice the ideal texture and flavor that fat provides (Li et al., 2014).

Flavored Milk in Schools

In 2010, the United States Government incorporated healthier foods into children's diets through the Healthy, Hungry-Free Kids Act (HHFKA) (Serdula et al., 1993). This Act established a nutritional standard for meals and beverages provided through school and after-

school programs to improve the health and nutrition of children to prevent obesity and other health-related issues by offering more fruit and vegetables, more whole grain products, and reducing the sugar and calorie content of foods products (Kenney et al., 2020). This Act was an update to current federal assistance programs like the National School Lunch Program (NSLP), School Breakfast Program (SBP) and Smart Snacks programs that are part of the Richard B. Russell National School Lunch Act (RBRNSLA) aimed to provide nutritious and well-balanced meals to millions of children in low-income households. Schools participating in the NSLP and SBP receive subsidies (cash or donated foods) from the programs for every meal served that follows the most recent Dietary Guidelines for Americans (Sipple et al., 2020). Federal subsidies are important for milk consumption, as four times as many students drank milk when provided for free compared to students in schools that required payment, along with children dislike spending their own money on milk products (Henry et al., 2015).

As part of the Dietary Guidelines at the time, participating schools were required to offer 1 cup of milk per day to students that may be either low-fat (1% or less, unflavored) or fat-free (unflavored or flavored), meaning schools could no longer serve 2% fat plain milk or low-fat flavored milk if they wanted to participate in the federally funded school meal program (Code of Federal Regulations, 2019). The removal of 2% plain milk and 1% flavored milk was meant to reduce the content of saturated fats, sugar and calorie content of school lunch menus. Some school districts like Washington D.C. and San Francisco even banned the sale of flavored milk to reduce the risk of child obesity due to added sugar and excess calories (Sieff, 2011; Tucker, 2017). Food behavior scientists argued that children are reactant to paternalistic policies (forced behavior) and a removal of flavored milk from school menus and forced consumption of plain milk may lead to a decrease of overall milk consumption (Goto et al., 2013; Hanks et al., 2014).

Removing flavored milk from school cafeteria resulted in elementary school aged children (ages 5 to 13 y) reaching for less milk and consuming less of the plain milk they did take because children preferred the taste of flavored milk (De Pelsmaeker et al., 2013; Hanks et al., 2014). The removal of flavored milk caused a 35% decrease in elementary student milk consumption and a 23% drop in the amount of milk consumed, as well as loss of essential nutrients (MilkPEP, 2017). The decline in milk consumption in US schools, along with a decrease in school lunch program participation forced the USDA to initiate an interim rule called the "Flexibilities for Milk, Whole Grains, and Sodium Requirements" which allowed flexibilities to Child Nutrition Program menus to relieve challenges food operators faced due to strict nutrition guidelines from the HHFKA of 2010, which included the sale of flavored, fat-free milk in 2017, and the final rule implemented in 2019 allowed 1% fat-flavored milk be served in schools (Federal Register, 2017, Federal Register, 2018).

The reduced milk consumption and greater waste in plain milk in schools indicated that students were not used to drinking plain milk at lunchtime or have an overall preference for chocolate milk over plain milk (Henry et al. 2015). Hanks et al. (2014) found that the sale of white milk increased by 161.2 cartons per day, but total dairy milk sales decreased by 9.9% when chocolate milk was removed from school lunch menus, while 29.4% of all milk purchased was disposed of. In this study, children eventually transitioned to consuming plain milk and reduced their total sugar consumption, but overall calorie and nutrient content from milk decreased, as well as decreased total participation in school lunch programs as well, which could impact funding from federal assistance programs (Hanks et al., 2014). Flavored milk is crucial for child growth and development because it consists of essential nutrients and health benefits for children that should not be overlooked due to its added sugar and calorie content. The removal of

flavored milk from school lunches may reduce the total calories and added sugar consumed by children, but it may also lead to fewer students purchasing school lunches, less milk consumed overall, and lack of nutrient intake in growing children.

Calorie Reduced Flavored Milk

Calorie reduction in flavored milk can be achieved by reducing the fat content or added sugar content. Several reports have shown that children prefer foods with higher fat content (Johnson et al., 1991; Kern et al., 1993; Kildegaard and Løkke, 2011). Milkfat found in dairy products can affect the sensory properties of fluid milk, such as altering the mouthfeel, viscosity, aroma and flavor attributes (Phillips et al., 1995; Phillips and Barbano, 1997; Frøst et al., 2001; Chojnicka-Paszun et al., 2012; McCarthy et al., 2017). When measuring child preference and liking of low-fat and high-fat fluid milk, Kling et al. (2016) reported children rated the two milks similarly, with no difference in the distribution of liking and preference results. In this study, there was no difference in the distribution in liking and preference among boys and girls populations.

More recently, Keefer et al. (2022) reported higher overall child liking scores for 1% and 2% fat milks compared to skim for unflavored milk when visual cues were presented, and higher liking scores for 1% and 2% fat chocolate milk over skim milk. When visual cues were not presented, Keefer et al. (2022) found reported no differences in child liking, flavor, or mouthfeel for among skim, 1% and 2% fat unflavored milk. These studies were conducted with the absence of any other confounding factors (package, light exposure) on fluid milk flavor other than milkfat content. Keefer et al. (2022) also suggested that children would have a more positive perception of milk if they consumed milk with higher fat content, and schools should increase the fat content in milk served to increase positive attributes and promote consumption of milk

among children. For adults, liking and preference for fat content in milk was based on the sensory characteristics of the milk consumers typically choose (Bakke et al., 2016; McCarthy et al., 2017). The effect of milk fat typically consumed may not be evident with children because they have not established habits and preferences (Ventura and Worobey, 2013; Sipple et al., 2021).

Another method to reduce the total calories in flavored milk and dairy products is to reduce the added sugar (McCain et al., 2018). Overconsumption of sugar-sweetened foods and beverages is a major contributor to the increased rate of obesity and unhealthy weight gain over the last two decades among Americans (Dubowitz et al., 2008; Han and Powell, 2013; May et al., 2013). The *Dietary Guidelines for Americans* (2020) recommends added sugar should make up less than 10% of total energy intake per day starting at 2 years. Between 2017-2018, the average added sugar intake was 17 teaspoons for children and young adults aged 2 to 19 years, as well as for adults aged 20 and older (USDA-ARS, 2020).The National Health and Nutrition Examination Survey (NHANES) 2013-2016 data estimated the average energy intake from added sugar for Americans ages 1 year and older resulted in 266 calories, which equates to about 13% of total calories per day, with 24% of those calories coming from sugar sweetened beverages, 19% from dessert and sweet snacks, and 11% from coffee and tea (USDA-USDHHS, 2015), meaning most Americans consume diets that exceed the recommended *Dietary Guidelines*.

Approaches for sugar reduction in milk and flavored milk have been reviewed by McCain et al. (2018). This review covers the role of nutritive and nonnutritive sweeteners that can reduce the caloric content of dairy foods (e.g. milk, yogurt, ice cream) to improve the health of children and adults. This review also detailed methods for sugar reduction via lactose

hydrolysis, ultrafiltration, and direct reduction. A review by Wan et al. (2021) on sucrose reduction in yogurt revealed lactose hydrolysis was an effective method to reduce added sucrose content in dairy products, while using alternative sweeteners with fewer added sugar content and minimizing alteration of the sensory characteristics.

Studies have been conducted to evaluate sugar reduction in products and to measure consumer acceptance and liking. Li et al. (2015b) measured the effect of reducing sucrose content in chocolate milk with adults and children. The study confirmed that children and adults preferred chocolate milk with higher sucrose concentration, but also found that a maximum of a 30% sucrose reduction from 205 m*M* was accepted by both populations. Oliveira et al. (2016) reported no significant differences in overall liking scores for chocolate milks with different added sugar concentrations among adults up to a difference of 6.7%. Henry et al. (2016) reported that schoolchildren preferred reduced sugar chocolate milk over plain milk, but the total number of students consuming milk decreased as well. In Chollet et al. (2013), a sugar reduction in yogurt caused a decrease in overall liking, which is consistent with Markey et al. (2015) for overall liking of reduced sugar products.

Nonnutritive Sweeteners

Flavored milk can increase milk consumption among children and adults to provide beneficial nutrients and vitamins important for growth at all ages. Unfortunately, the added sugar content of flavored milk raises concerns among parents and schools because it is linked to obesity and health issues. Nonnutritive sweeteners (NNS) are impacting the food industry for their effectiveness in sweetening foods and their role in weight management and health benefits by reducing the added sugar and calorie content (McCain et al., 2018).

Body weight and adiposity are developed under conditions of a positive energy intake. Nonnutritive sweeteners are as sweet as or even sweeter than natural cane sucrose, but have a different caloric content and metabolism route, and thus may affect the energy balance and body weight differently compared to natural sucrose. Cohort studies on the association between low-energy sweeteners and human body weight have reported inconsistent results regarding the ability for nonnutritive sweeteners to reduce energy intake and body weight. Five studies reported a higher risk of obesity with the consumption of nonnutritive sweeteners (Berkey et al., 2004; Fowler et al., 2008; Nettleton et al., 2009; Duffey et al., 2012), while six studies reported a lower risk of obesity associated with the consumption of nonnutritive sweeteners (Ludwig et al., 2001; Striegel-Moore et al., 2006; Chen et al., 2009; Pan et al., 2013). The three largest studies in these cohort studies were reported as a pooled analysis, and reported evidence of lower weight gain when substituting sugar-sweetened beverages for diet beverages (Pan et al., 2013). Inconsistent results from these studies measuring impact of nonnutritive sweeteners on body weight are possibly due to other characteristics and behaviors of individuals participating in the studies. Anthropometric studies measuring the body weights of individuals who consume nonnutritive sweeteners versus sugar-sweetened beverages show significant differences in body weights in favor of nonnutritive sweeteners (Tordoff and Alleva, 1990; Blackburn et al., 1997; Raben et al., 2002; Reid et al., 2007), although many studies had differences that were not reported as statistically significant in body weight (Kanders et al., 1988; Raben et al., 2002; Reid et al., 2007; Njike et al., 2011; Tate et al., 2012). There is considerable evidence that nonnutritive sweeteners are helpful in reducing relative body weight and energy intake in humans, with no evidence of nonnutritive sweeteners increasing either body weight nor energy intake (Pang et al., 2020).

Allulose

Allulose (D-Allulose, D-psicose) is a naturally occurring low energy monosaccharide sugar that is approximately 70% relative sweetness of sucrose and 0.4 calories per gram (Jiang et al, 2020). Found in wheat, corn, beet sugar and fruits, allulose is a C3 epimer of fructose (contains a hydrogen atom and a hydroxyl group in the 3rd carbon position) of fructose (Jiang et al., 2020. Approved as "generally recognized as safe" (GRAS) by the FDA in the USA in 2019, allulose is effective in suppressing blood-glucose and insulin elevations after eating and postprandial blood-glucose elevation with borderline diabetic subjects (Franchi et al., 2021; Iida et al., 2008; Hayashi et al., 2010). Hayashi et al. (2010) found no significant differences in insulin levels nor AUC values when comparing postprandial glucose levels in diabetic adults after consuming tea with zero or 5 grams of allulose with a standard meal. Studies have shown that majority of allullose is rapidly eliminated from the body via urine or feces without being metabolized, with 2% of consumed allulose absorbed by the small intestine (Whistler et al., 1974; Hossain et al., 2015; Maeng et al., 2019). Similar to erythritol, allulose is not metabolized by human gastric fluid, thus remains highly stable through digestion and is not involved in glucose related metabolism nor contributes to hepatic energy production (Iwasaki et al., 2018; Maeng et al., 2019). Allulose suppresses hepatic lipogenic enzyme activity by lowering fatty acid synthase and glucose 6-phosphate dehydrogenase activities in the liver, which aids in lower abdominal fat accumulation and increased body fat oxidation (Matsuo et al., 2001; Han et al., 2016, 2018b). This sweetener demonstrated strong anti-hyperlipidemic, anti-hyperglycemic effects that are important for consumers suffering from obesity and type 2 diabetes (Hossain et al., 2015; Han et al., 2018b; Van Laar et al., 2020).

Drawbacks to using allulose consists of a maximum intake capacity and gastrointestinal effects. Studies associate a digestive tolerance with D-allulose consumption to a maximum single dose of D-allulose of 0.4g allulose/kg*Body Weight, and a maximum daily intake of 0.9g allulose.kg*BW (Han et al., 2018a). Gastrointestinal symptoms that occurred when consuming past the maximum intake include abdominal distention, abdominal pain, and diarrhea being the most common symptom occurring (Han et al., 2018a).

During processing and application, allulose is described as highly soluble, improves gelling behavior, mouthfeel and browning appearance (Oshima et al., 2006; Sun et al., 2008). Sun et al. (2008) found that allulose can increase the foaming and browning properties when combined with egg white protein to form butter cookies compared to sucrose and fructose. Sun et al. (2008) also determined that the use of allulose did not affect the cook loss of the butter cookies, nor and significantly contributed to a color change on the cookie crust via nonenzymatic browning reaction. The butter cookies that contained allulose also possessed the highest antioxidant capacity in all tested cookies when measured for radical scavenging activity and ferric reducing power (Sun et al., 2008). Allulose is especially effective in baking and confectionary applications to capitalize on improved sweetness, mouthfeel, and browning appearance, as well in beverage form due to being processed into a crystalline and syrup form (Sun et al., 2008).

Ates et al. (2020) found that soy protein gels sweetened with allulose had higher crystallization tendency compared to sucrose sweetened soy protein gels since allulose has a lower water binding ability. Soy protein gels mixed with allulose increased the gel strength compared to sucrose, thus affecting the digestive behavior of the product, which could be beneficial for designing low-calorie confectionary products that promote satiety sensations (Ates

et al., 2021a; Ates et al., 2021b; Xia et al., 2021). Kimoto-Nira et al. (2017) found that allulose can suppress acid production in certain dairy lactic acid bacteria in yogurt without affecting their probiotic activity and could be useful to develop probiotic dairy products from dairy probiotics candidate strains. Zeng et al. (2013) used allulose and fructose to modify bovine β-lactoglobulin (β-Lg) through Maillard reaction and results showed that allulose was more effective in producing Maillard reaction compared to fructose. The allulose modified β-Lg sample had more polymeric compounds, higher antioxidant activity, but a lower thermal stability compared to fructose modified β-Lg (Zeng et al., 2013).

Monk Fruit

Monk fruit is a natural, nonnutritive sweetener extracted from *Siraitia grosvenorii*, or *Luo Han Gao*, and is 150-200 times sweeter than table sugar (Kim and Kinghorn, 2002). Luo han guo is a small round fruit of the Cucurbaitabacae family and native to the Guangxi province in southern China. Monk fruit consists of mogrosides, which are triterpene glycosides that account for the inherent sweetening property, as well as essential oils, saccharides, proteins and vitamins responsible for the fruit's nutritional content and flavor. The mogrosides responsible for the strong sweetness property of the fruit consist of mogroside IV (392x sweeter than sucrose), mogroside V (425x), siamenoside I (563x) and 11-oxo-mogroside V (84x), and are significantly sweeter than sucrose (Kim et al., 2015; Pandey and Chauhan, 2020).

The potential for monk fruit as a nonnutritive sweetener has not been fully developed yet as a sugar alternative. It is an effective part of dietary changes to fight and prevent health related diseases due to its low calorie content and high sweetness potency. Studies have shown that monk fruit extract and its inherent natural compounds are nontoxic and do not show adverse effects to the human body post-consumption (Li et al., 2014; Liu et al., 2016). Mogroside IV, one

of the mogrosides found in monk fruit, has been found to exert anti-hyperglycemic effects and regulation of blood sugar levels in diabetic patients (Liu et al., 2016).

Monk fruit extract exhibits antihyperglycemic, cancer prevention, and anti-diabetic effects in animal models that could carry-over to human results (Liu et al., 2016). In studies involving diabetic rats as subjects, monk fruit exhibited increased insulin response and total pancreatic insulin content, while also having reduced plasma glucose levels and thiobarbituric acid-reactive substances in the liver and plasma, suggesting monk fruit may prevent and minimize complications with Type 2 diabetes while providing a favorable sweet taste characteristic (Suzuki et al., 2007). Another study on mogroside IV exhibited anti-cancer effects by inhibiting the proliferation of colorectal and throat cancer cells when applied *in vitro* and *in vivo* in cultures and xenografted tumor mice models containing cancer cells (Liu et al., 2016). The mogroside IV-treated mice demonstrated a significant increase in the number of apoptotic bodies compared to the control mice group, suggesting that mogroside IV suppresses cancer tumor growth by inducing apoptosis in cells (Liu et al., 2016). The demand for nonnutritive sweeteners has made monk fruit a popular commodity in the food and beverage industries.

Monk fruit is harvested before ripening occurs, when the fruit is in an unripe state, to allow it to fully ripen during storage. The sweetness levels of monk fruit vary depending on the ripening process and the amount of mogroside V present in the fruit. The fruit is ripened during storage to prevent infection of fruit flies and insects that can induce decay (EFSA Panel on Food Additives and Flavourings (FAF) et al., 2019). As the fruit ripens during storage and reaches peak sweetness, the extraction process begins. The peels and seeds are first removed, and the fruit is smashed in a paste, where juices are released from the fruit matrix (Fry, 2012). The juice can be acidified to increase the pH to prevent gelling during the concentration process. Off-

flavors are removed, like sulfurous materials or sulfur-containing amino-acid materials, by treatment using enzymes, precipitant agents, adsorbents, and ion exchange resins. The concentration of monk fruit extract is completed by a multi-effect evaporator that uses steam to remove water from the fruit extract, as well as remove undesirable flavors (Shivani et al., 2021). Vacuum drying is another method to harvest monk fruit extract that yields a high amount of major glycosides (Pandey and Chauhan, 2019). Shi et al. (2009) patented a process to develop monk fruit syrup and jams using the peels and pulp of the monk fruit that resulted in a sugar-free, low calorie and low glycemic index product. Shi et al. (2009) boiled the monk fruit to extract components from the peel and pulp, then used pectinase to digest pectin that naturally found in monk fruit to prevent dilution, layering and ensure stability during storage. The remaining extract liquid is filtered to remove small pieces of monk fruit using different mesh filters and prevent bacterial growth (Shi et al., 2009).

Due to its high cost, low production quantity, and difficulty in harvesting, monk fruit has not been fully utilized in food production. The fruit is mainly found in the mountainous terrain of Southern China and requires extensive labor to grow and harvest the crop. After harvest, the fruit is transported to a processing facility to remove water and pulp to concentrate the mogrosides present in the fruit. This refining process, along with shipping across the world, causes monk fruit extract to be expensive and not suitable in the food market for certain companies.

Monk fruit has been approved as Generally Recognized as Safe (GRAS) by the U.S. Food and Drug Administration in a majority of its forms to be used in food production. A research study by the European Food Safety Authority found that monk fruit does not induce chromosomal damage, nor any effects of toxicity on the reproductive or developmental processes in rats (EFSA Panel on Food Additives and Flavourings (FAF) et al., 2019). There were effects

on the liver and testis of the rats during the experiment, which could be the result of adaptive responses, but significant enough where the panel could not rule out the findings. This study was conducted in a 90-day trial, and further research must be conducted to conclude the findings in this experiment.

Literature reports suggest a wide potential for utilization of monk fruit extract in food and beverage industries (Shi et al., 2009; Heine, 2017). Shi et al., (2009) found that monk fruit extract could be combined with sugar alcohols (maltitol) to create a sugar free syrup, or mixed with sugar alcohol, calcium citrate, citric acid, and pectin to form a monk fruit extract jam (Shi et al., 2009). Heine (2017) developed a chocolate product that contains a monk fruit blend monk fruit fiber and extract that is low in fat and complex carbohydrates and is beneficial for consumers who are insulin resistance or suffer from diabetes. Quinlan and Zhou (2017) created a non-caloric sweetener composed of monk fruit extract and *stevia rebaudiana* (Rebaudiosides A and B) that is useful for non-caloric replacements of sugar in foods and beverages with reduced levels of perceived bitterness.

Li et al. (2015a) found that chocolate milk with 25% monk fruit with sucrose (9.39 g/serving of sucrose and 10.8 mg/L monk fruit) were acceptable among children and adults, but not accepted at higher concentrations of monk fruit. Buchilina and Aryana (2021) applied varying levels of monk fruit to camel milk-based drinking yogurt to examine the influence of monk fruit on the physiochemical properties and microbiological count of camel milk drinking yogurt. The addition of the monk fruit influenced the pH, viscosity, and color of the yogurt, and did not affect the growth of microbiological cultures. Ban et al. (2020) found that rats who consumed monk fruit sweetened yogurt improved blood glucose regulation and a significant decrease in insulin resistance and glycosylated hemoglobin compared to rats who did not

consume the monk fruit sweetened yogurt. This study exemplifies that monk fruit extract can be a beneficial sweetener substitution for people with type 2 diabetes to delay the progression of diabetes and associated complications (Ban et al., 2020).

Sucralose

Sucralose is a non-caloric high intensity sweetener that is approximately 350-600 times sweeter than sucrose (Ma et al., 2010). Derived from sugar, sucralose is produced by replacing three hydroxyl groups of a sucrose molecule and replacing it with three chlorine atoms. Approved by the FDA as sweetener in foods generally in 1998, sucralose is widely applicable in food, beverage and pharmaceutical products around the world due to its solubility in ethanol, methanol and water and is most commonly found in sodas, ice cream, frozen desserts, chewing gum baked goods and fried goods (Li et al., 2010; Schiffman and Rother, 2013).

Sucralose is not readily absorbed by the intestinal tract, with the majority of the sweetener excreted in feces and ~5% excreted via urine, resulting in no calories digested in the body (Sims et al., 2000). In human study of sucralose metabolism, eight male subjects were administered a single oral dose (1 mg/kg, 100μCi) of radioactive sucralose, and an average of 92.8% of the sweetener was excreted within 5 days, with the radiolabeled sucralose present in the feces being essentially unchanged (Roberts et al., 2000). Other studies have shown that an average of 78.3% of sucralose is eliminated via digestion tube and 14.5% via urine (Rodero et al., 2009). Goldsmith (2000) evaluated the acute toxicity of sucralose in mice by administering 1%, 2%, and 5% sucralose solutions. 1% and 2% solutions did not show any significant impacts on health, 5% concentration of sucralose (in rats = 2794-6406 mg/kg/day) showed diminished food consumption, weight gain, deficient food conversion and changes in organ weights, alterations in the cecum (Goldsmith, 2000). Research studies administering sucralose have

showed no evidence of gut microflora adaptation or mammalian metabolizing systems, as well as no effect on body weight, glycaemic control, and insulin resistance (Sims et al., 2000; Ma et al., 2010; Sylvetsky et al., 2011; Thomson et al., 2019). Sucralose studies in school aged children reduced weight gain and fat accumulation when comparing sugar-free beverages sweetened with sucralose to sugar-sweetened beverages (de Ruyter et al., 2012).

Morais et al. (2014) analyzed the ideal and relative sweetness of prebiotic chocolate milk dessert at 8% sucrose against sucralose and other nonnutritive sweeteners. Morais et al. (2014) found that sucralose has the second highest relative sweetness in the chocolate milk dessert after neotame at a sweetening power of 500 or have a sweetness concentration equivalence of 0.016% to 8% sucrose in prebiotic chocolate milk dessert products. Markey et al. (2015) determined that cranberry and raspberry juice sweetened with sucralose exhibited a different sweetness profile (length of impact of sweet taste) compared to the juice sweetened with sucrose.

Parker et al. (2018) used descriptive analysis, temporal dominance of sensations, and consumer acceptance to evaluate ready-to-mix (RTM) vanilla protein beverages sweetened with sucralose (130.7 mg/L RTM beverage) and other nutritive and nonnutritive sweeteners at equisweet intensity. By descriptive analysis, the sucralose-sweetened RTM vanilla beverage displayed bitter and metallic tastes, and Temporal Dominance of Sensations results showed a dominant initial sweet taste, followed by sweet aromatic and cardboard flavor (Parker et al., 2018). Cluster analysis of consumer acceptance results in Parker et al. (2018) determined two distinct segments of consumers of RTM vanilla protein beverages. Segment 1 consumers (n=75) were reported as less frequent consumers of sucralose compared to Segment 2 consumers (n=75) who scored primed sucralose samples higher than the unprimed samples, suggesting that Segment 1 consumers are more label conscious, while Segment 2 are primarily influenced by

flavor. Both segments of consumers in Parker et al. (2018) scored the sucralose sweetened protein beverages the lowest in concept liking, but Segment 2 consumers scored sucralose beverages the highest in flavor liking, sweetness liking, and vanilla flavor liking while Segment 1 consumers scored the sucralose beverage highest in flavor liking when unprimed. Parker et al. (2018) concluded that the sucralose sweetened sample scored at parity with reduced-sugar blends beverages containing fructose for overall liking and flavor liking across all consumers when unprimed.

Rodriguez Furlán and Campderrós (2017) evaluated the effect of replacing sugar with sucralose and stevia in starch-based dairy desserts and measuring the sensory, physiochemical, and textural properties. Using a 50%-50% blend of stevia and sucralose, this sample obtained the highest sensorial acceptability and was not statistically significantly different compared to the 100% sucrose dessert sample (n=52), all while tying for the highest values of viscosity and consistency factor with the 100% stevia sample (Rodriguez Furlán and Campderrós, 2017). This study also combined stevia, sucralose, and inulin (50% stevia, 50% sucralose, 2.5% inulin) and found it to have no significant sensory differences other than texture and was preferred when compared against a reduced-sugar commercial dairy dessert sample (Rodriguez Furlán and Campderrós, 2017).

Palla et al. (2020) examined a complete replacement of sucrose with sucralose in low calorie ice cream and found that the sucralose-sweetened ice cream had a significant decrease in mouthfeel, body and textural feel compared to the sucrose control ice cream. The sweetness level of the sucralose ice cream at 300 ppm was close to the sweetness level of the sucrose control ice cream at 15% sucrose, but lacked similar mouthfeel (Palla et al., 2020). To achieve a similar mouthfeel texture, sorbitol was incorporated to the sucralose-sweetened ice cream and at 3%

sorbitol, the sample was the most comparable to the sucrose control ice cream based on viscosity, percent overrun, penetration value, sensory evaluation and melting rate (Palla et al., 2020). This study concluded that sucrose replacement with sucralose and sorbitol significantly affected the sensory attributes of ice cream, and a 300-ppm sucralose with 2% sorbitol was highly acceptable among consumers when compared with the sucrose control low-calorie ice cream (Palla et al., 2020). The replacement of sucrose with sucralose and sorbitol provided a calorie reduction of 60.71 Kcal/100 grams (Palla et al., 2020).

Stevia

Steviol glycosides are the chemical compounds responsible for the sweet taste in the South American plant *stevia rebaudiana* (Cardello et al., 1999; Kim and Kinghorn, 2002; Pang et al., 2020). Stevia extract became available in the US in the 1990s as a dietary supplement in health food stores and was known to have a sweet licorice flavor with a bitter aftertaste. It was approved as GRAS by the FDA for specific conditions of use in 2018 (FDA, 2018). There are 11 different types of steviol glycosides, but the most common type of steviol glycosides used in food processing are stevioside and Rebaudioside A, which have sweetening power of 150-300 and 200-400 times relative to sucrose, respectively (Ashwell, 2015).

Stevia is processed into stevia leaf extract similar to sugar and vanilla extract, where the raw plant material is harvested, dried, and then steeped in hot water (Ashwell, 2015). The liquid extract is filtered and purified with water or combined with food-grade alcohol. Stevia leaf extract is normally found at 95% steviol glycosides to meet US and European regulatory approvals and safety standards (Ashwell, 2015).

Steviol glycoside cannot be broken down by the digestive enzymes and acids present in the upper gastrointestinal tract (Wingard et al., 1980). The steviol glycosides are degraded by

microbiota in the colon by cleaving the glycosidic linkages forming steviol, steviolbioside, and glucose (Wingard et al., 1980; Gardana et al., 2003; Pang et al., 2020). The steviolbioside is converted into steviol, and the glucose is utilized by the colonic bacteria or absorbed, metabolized or excreted into the air as carbon dioxide and water (Carakostas et al., 2008; Pang et al., 2020). The steviol is then absorbed by the portal vein and metabolized by the liver to form steviol glucuronide and excreted via urine. The Adequate Daily Intake for steviol equivalents is 4mg/ kg of body weight per day (Pang et al., 2020).

In starch-based dairy desserts, stevia produced the highest viscosity and consistency along with a 50%-50% blend of stevia and sucralose when compared with sucrose (Rodriguez Furlán and Campderrós, 2017). When combined with sucralose, stevia in the dairy dessert had the highest sensory acceptability and was not significantly different from the control sucrose dairy dessert (Rodriguez Furlán and Campderrós, 2017). Parker et al. (2018) measured consumer acceptability when stevia was used as part of a natural sweetener blend in protein beverages. Parker et al. (2018) found that specific clusters of consumers preferred naturally sweetened beverages when primed, but flavor-driven consumers preferred sucralose-sweetened beverages over stevia. Of the reduced sugar blends in protein beverages in this study, sweetener blend 4 (25% stevia/25% monk fruit/50% fructose) and blend 7 (25% stevia/75% monk fruit) were very similar in temporal sensory properties to the sucrose control beverage (Parker et al., 2018).

Alizadeh (2014) measured the replacement of sucrose with stevia at varying concentrations in fruit-based milk shakes and its effect on sensory and physiochemical properties. The substitution of sucrose with stevia produced no remarkable effect on the pH, acidity, and vitamin C content of the beverages, and the total soluble solids decreased in proportion to the reduction of sucrose content (Alizadeh, 2014). Among the samples containing

stevia, sample TD (25% sucrose, 75% stevia) had the highest mean liking score followed by sample TB (75% sucrose, 25% stevia), TC (50% sucrose, 50% stevia), and TE (100% stevia) (Alizadeh, 2014). Alizadeh (2014) recommended the ideal ratio in beverage for stevia was 25% sucrose and 75% stevia, and had no adverse impact of physiochemical properties, but sensory properties are affected by the rate of stevia addition.

Li et al. (2015a) evaluated the effect of substituting stevia and monk fruit for sucrose in skim chocolate milk with young adults and children. This study found that chocolate milk solely sweetened by stevia were less acceptable compared to the sucrose control beverage by young adults. Chocolate milks MK 25 (39.7 g/L sucrose and 46 mg/L monk fruit) and STV 25 (39.7g/L sucrose and 30 mg/L stevia) were acceptable among both young adults and children (Li et al., 2015a). Homayouni Rad et al. (2012) assessed the impact of chocolate milk by replacing sucrose with stevia and inulin. Homayouni Rad et al. (2012) found that sucrose replacement with stevia caused significant increase in precipitation and significant decrease in viscosity. The chocolate milk sample containing 100% stevia with 6% inulin produced a viscosity with no significant difference to the sucrose control chocolate milk. This study concluded that stevia sweetened chocolate milk can be acceptable among consumers with the improved physical properties from inulin (Homayouni Rad et al., 2012). Bordi et al. (2016) used stevia to reduce the sugar content of chocolate milk by 35% using 150 ppm of Reb A stevia without impacting overall liking.

Magnitude Estimation Scaling

Magnitude estimation scaling (MES) is a popular technique in psychophysical science to measure the intensity of a sensory stimuli by assigning numbers to the perceived magnitude of the stimulus (Moskowitz, 1970; Lawless and Heymann, 2010a). Respondents designate an algebraically positive number to a particular stimuli or sensation that reflects an intensity that is

perceived by the respondent. Lavenka and Kamen (1994) applied magnitude estimation to the acceptance of food where the numbers designated to a stimuli's intensity should be proportional to the ratios of sensation magnitudes that have been experienced. MES can estimate the equivalent sweetness, also known as the *isosweet concentration*, of sweeteners at known concentrations, compared to a standard modulus (sucrose at a standard concentration) (Lavenka and Kamen, 1994; Lawless and Heymann, 2010a). The concentrations of the sweeteners are diluted at constant ratios, known as step factors, to describe the apparent differences in magnitude of the sample. MES provides a standard stimulus to the respondent as a reference, also called a "modulus" (Lawless and Heymann, 2010a). The standard stimulus is assigned a fixed value, somewhere near the middle of the intensity range, so respondents can determine stimuli intensity of samples with stimuli at unknown concentrations with improved accuracy.

The validity of magnitude estimation scaling is based on Steven's Power Law, which posits that response to a stimulus is a power function of intensity. Steven's Power Law is a relationship between an increased intensity or strength of a physical stimulus and the perceived magnitude increase in the sensation caused by the stimulus. R is the intensity or strength of the stimulus in physical units, S is the magnitude of the sensation evoked by the stimulus, n is the exponent that depends on the type of sensation modality, and k is the proportionality constant that depends on the units used.

$$S = k R^{n}$$

This relationship originates from the Weber-Fechner law, which is based on a logarithmic relationship between a stimulus and sensations. Weber's law quantifies the perception of change in a given stimulus, and that just noticeable difference (JND) or change in stimulus is

proportional to the original stimulus (Lawless and Heymann, 2010a). Fechner's law states that the relationship between the stimulus and perceived sensation is logarithmic, meaning if the stimulus varies as a geometric progression (multiplied by a fixed factor), the corresponding perception of the stimulus will be an arithmetic progression (Lawless and Heymann, 2010a).

Iso-Sweet Concentration of Alternative Sweeteners

There have been multiple studies that used magnitude estimation scaling to determine sweetness equivalence of sucrose to other nutritive and nonnutritive sweeteners (Stone and Oliver, 1969; Cardoso and Bolini, 2007; Li et al., 2015a; b; Parker et al., 2018; Wagoner et al., 2018). Wee et al. (2018) used magnitude estimation scaling to determine the equivalent sweetness concentration of nutritive saccharide, nutritive polyol, and nonnutritive natural sweeteners to sucrose at 10% w/v sweetness intensity. Slope of the linear fit model between log sweetness intensity and log concentration was highest for allulose (1.41), followed by stevia (0.71), monk fruit (0.7), and sucralose (0.65). Wee et al. (2018) also stated these sweeteners exhibited a sigmoidal dose-response curve between sweetener concentration and perceived intensity due to the binding kinetics of sweetener molecules to taste receptors, which plateaued at higher concentrations when the receptors become saturated. Using magnitude estimation scaling, Antenucci and Hayes (2015) found that nonnutritive sweeteners have a lower maximal sweetness intensity than sucrose due to mixture suppression of sweetness by bitter or metallic sensations at increasing concentrations. The bitterness found in stevia, monk fruit and sucralose provides a ceiling for maximum sweetness, but can also reverse the slope of the power function at higher concentrations, which have been seen in glycoside rebaudioside (Hellfritsch et al., 2012). Dose-response functions for sucralose and stevia Rebaudioside A were shifted left compared to

sucrose, exhibiting a higher potency than sucrose, but bitter taste increased with concentration to equal or surpass perceived sweetness (Antenucci and Hayes, 2015).

Reis et al. (2011) used magnitude estimation scaling to evaluate the sweetness equivalence of sweeteners and sweetener blends of sucrose, aspartame, acesulfame-K, sucralose, cyclamate, stevia and saccharin in strawberry yogurt. The sweetener blends presented similar logarithmic curves to the sucrose control sample due to synergistic effects to produce a superior equivalent sweetness with more pleasant flavors. When comparing power function slope values of sweeteners in cold tea at 8.3% w/w sucrose in Cardoso et al. (2004), the angular coefficient (n) values were higher compared to the coefficient values with sweeteners in Reis et al. (2011), suggesting an influence of the product's characteristics on the sweetness perception.

In Parker et al. (2018), ready-to-mix (RTM) protein beverages sweetened with sucralose, fructose, stevia and monk fruit were measured for sweetness equivalence to 7% sucrose solution. Sucralose was the most potent sweetener, followed by monk fruit, stevia, and fructose. Parker et al. (2018) found no difference in iso-sweetness concentration for the sweeteners when using different protein levels (15 vs 25g/360 mL serving size) and whey protein isolate (WPI) type (anion exchange vs micro filtered). As such, the glycomacropeptide in microfiltered WPI does not affect sweet taste perception.

Li et al. (2014) found that parents preferred natural nonnutritive and noncaloric sweeteners over artificial sweeteners in regards to sugar reduced products. Li et al. (2015a) examined the adult acceptance of skim chocolate milk with 25%, 50%, 75% and 100% sugar reduction that contained stevia and monk fruit extract. No difference in overall liking, sweetness and aftertaste liking was observed among the sucrose control, stevia 25% and monk fruit 25% milks, and these milks received the highest liking scores. Complete sucrose replacement with

stevia and monk fruit received the lowest scores for overall liking, flavor, sweetness, cocoa flavor, and aftertaste due to the presence of bitter and metallic tastes. This result is consistent with (Reis et al., 2011) that reported stevia and acesulfame-K presented high bitter tastes in the yogurt as the sole sweetener, showing these nonnutritive sweeteners alone could not produce sweetness sensations in strawberry yogurt that are identical to sucrose in aqueous solutions (Reis et al., 2011). Blends of nutritive and nonnutritive sweeteners can be used to maximize sweetness without the presence of persistent off-flavors like bitter and metallic tastes that intensifies as increasing concentration (Reis et al., 2011).

Li et al. (2015a) evaluated consumer acceptability of reduced sucrose chocolate milk with stevia and monk fruit extract with school-aged children. No differences were observed in overall liking, sweet taste, and chocolate liking for the chocolate milks. There was also no difference in liking, confirming that chocolate milk sweetened with stevia and monk fruit extract blends were acceptable among children (Li et al., 2015a). Castillo et al. (2005) reported that milk sweetened with xylitol was acceptable among children. Aspartame-sweetened chocolate milk had similar intake to sucrose-sweetened chocolate milk with school aged children (Wilson, 2000). Chadha et al. (2022) investigated the effects of adding different natural sweeteners to yogurt and the impact on the temporal sensory profile and consumer acceptance of the yogurt. Using sucrose as a control sample (9 grams sucrose/100 g yogurt), Chadha et al. (2022) determined the iso-sweet concentrations of xylitol (10g/100g), stevia (0.15g/100g), and monk fruit (0.15g/100g) in the unsweetened yogurt base.

Lactose Hydrolysis

Lactose is a naturally occurring disaccharide that is exclusively found in the milk of mammals (Schaafsma, 2008). The carbohydrate forms 54% of the total non-fat milk solids and

provides 30% of the energy of milk (Saxelin et al., 2003). It is not as sweet as sucrose, fructose or glucose, with a relative sweetness of about 0.2-0.4 when compared to sucrose (Johnson and Conforti, 2003; Hobbs, 2009; Tiefenbacher, 2017). Lactose is 9.4 times less sweet than standard sucrose, but when hydrolyzed into glucose and galactose, the combination of monosaccharides is 1.5 less sweet than sucrose (McCain et al., 2018). Lactose is formed when a D-glucose and a D-galactose molecule are joined at the β-1,4-glycosidic linkage into two isomeric forms, α- and β-lactose depending on the steric configuration of the hydroxyl group of the C-1 of glucose (Johnson and Conforti, 2003). Lactose is the most abundant milk solid particle and is important in probiotic bacteria growth in the gut (Haug et al., 2007; Fernandez-Raudales et al., 2012; Aslam et al., 2020). When hydrolyzed, lactose is broken down into monosaccharides glucose and galactose, which both have a higher relative sweetness compared to lactose, and do not have bitter or metallic tastes and aftertastes like artificial sweeteners (Li et al., 2014; Tiefenbacher, 2017; Roy et al., 2020).

First applied to dairy products in 1950 and commercially available in 1970, lactase is an important enzyme to accelerate the processing time and improve the taste and texture of dairy products. Hydrolysis of 70% of lactose in milk has shown to increase the sweetness in milk or yogurt, resulting in the same degree of sweet taste as adding 2% of sugar (Zadow, 1986; Mahoney, 1998; Adhikari et al., 2010). Adhikari et al. (2010) found lactose-free milk to have higher intensities of sweet taste, cooked and processed flavors, and chalkiness texture when compared to regular milk. Lactose-free milk is perceived as sweeter than regular milk due to conversion of lactose into glucose and galactose (Chapman et al., 2001; Adhikari et al., 2010). Li et al. (2015b) measured the effect of adding lactase to chocolate milk and its acceptability with young adults and children. Hydrolyzed skim chocolate milk without added sugar reported a

higher sweetness score (1.5 out of 15) compared to chocolate milk with no hydrolyzed lactose (0 out of 15) with descriptive analysis. Hydrolyzed chocolate milk with permeate and 10 g of lactose matched the lowest acceptable sweetness of chocolate milk (150 m*M* sucrose) but sodium, potassium, and organic acids found in permeate invoked a salty taste not suitable for consumption (Frankowski et al., 2014; Li et al., 2015b). Consumer test results showed no difference in sweetness liking between chocolate milk with 10g/100mL hydrolyzed lactose and 150 m*M* sucrose milk (Li et al., 2015b). These results suggest lactose hydrolysis cannot be the sole sweetening agent in chocolate milk due to the amount of lactose required to match the sweetness of sucrose, and the presence of bitter taste due to cocoa. Hydrolysis of lactose in yogurt has also led to sweeter yogurt without added sugar (Engel, 1973; Tamime and Deeth, 1980). Abbasi and Saeedabadian (2015) found that lactose hydrolyzed milk-produced ice cream with higher viscosity, decreased freezing point, while the surface tension and density was consistent with ice cream made with regular milk. This study also found that low sugar and low lactose ice creams had lower hardness and sweetness intensities, but increased resistance to melting. In Panesar et al. (2010), the quality of ice cream significantly improved with lactose hydrolysis by preventing the crystallization of lactose and reducing the sandy texture. Lactose hydrolysis is beneficial to food processors to accelerate the ripening and acidification processes in cheese and yogurt because glucose and galactose ferment more easily, thus reducing the overall time to achieve the preferred pH (Panesar et al., 2010; Saqib et al., 2017). Rizzo et al. (2020) found that the majority of lactose-free milk consumers preferred the higher sweet taste compared to regular milk and disliked the high viscosity and high eggy flavor intensity (due to ultra-pasteurization). This study also reported that consumers of lactose-free milk prioritized the

price of the product when purchasing, since lactose-free milk is usually more expensive than regular fluid milk.

The hydrolysis of lactose in milk is most commonly processed using the enzyme β-D-galactosidase, also known as lactase, that can be isolated from plants, animal organs, bacteria, yeasts, and fungi (Richmond et al., 1981; Panesar et al., 2010). Lactose hydrolysis of milk can be accomplished by the addition of β-D-galactosidase directly into milk and storing at 4°C for a set amount of time or overnight, depending on opportunity for microbial growth (Park et al., 1979; Zadow, 1986). The hydrolyzed milk is then thermally processed to inactivate the lactase enzyme (Harju et al., 2012). Another method is to add a sterile lactase enzyme into UHT milk before packaging, where the enzyme could hydrolyze at room temperature for a longer time (Dahlqvist et al. 1977). This method allowed for 1% of the enzyme to be used compared to non-sterile hydrolysis conditions. In heat-treated processing conditions, the addition of lactase causes increased Maillard browning due to the increase in reducing sugars (Mendoza et al., 2005). The higher concentration and reactivity of monosaccharides from lactose hydrolysis results in greater levels of Maillard browning that can form by-products that include potential off flavors (Evangelisti et al., 1999; Braekman et al., 2001; Messia et al., 2007). Other methods for lactose hydrolysis include membrane recovery of soluble enzymes and immobilized systems which are more economical, efficient, and amenable to large scale continuous operations (Shukla and Wierzbicki, 1975; Zadow, 1986). Lactose hydrolysis is a viable option for sugar reduction while retaining sweet taste intensity that consumers will enjoy.

Impacts of Food Matrix on Flavor and Sweet Taste

The intensity of volatile and flavor compounds detected by consumers can be altered due to the composition of the food matrix. Depending on the molecular structure, compounds can

react differently based on the presence of lipids, proteins, and carbohydrates. The intensity of the flavor perceived by the consumer depends on the flavor retention and release of the specific compounds (Ammari and Schroen, 2018). Flavor retention and release occurs when flavor molecules are released from food in the mouth and the volatile flavor compounds pass into the olfactory system. Flavors are affected by intrinsic chemical properties of the flavor, the composition of the medium, and the environmental conditions (Ammari & Schroen, 2018).

Lipids in dairy products have been observed to contain high-saturated fatty acid levels, which can lead to increased low-density lipoprotein cholesterol (LDL), which increases the risk of cardiovascular diseases. These macronutrients can impact flavor by binding volatile aroma molecules to prevent release into the air (Guichard, 2002). Lipids have been shown to induce significant retention in hydrophobic flavor compounds that can result in noticeable differences in perception compared to water (Guichard, 2002). Roberts et al. (2003) showed that solid phase lipids (hydrogenated palm fat) increased flavor release of volatile compounds compared to liquid phase lipids (coconut oil, milk fat). When the solid phase lipid (hydrogenated palm fat) was heated to contain zero percent solid fat content, there was no difference in flavor release from that observed in milk fat emulsions. The difference in flavor perception is caused by the polarity of the matrix and flavor compounds. Substances of low polarity (long chain fatty acids) have lower flavor thresholds in oils and higher thresholds in water (Forss, 1969). When flavor compounds bind to solvent molecules, it reduces volatilization out of the matrix, thus reducing the perception of the flavor. Compounds that are polar will bind to water, while nonpolar compounds will bind to oil-based matrices. The practical application is that a full fat food matrix is likely to be distinct in flavor from a reduced or fat free version of the same food matrix.

Lipids can also affect the ability to sense flavor compounds on the tongue. Oil can coat the tongue and mix with saliva in the mouth, which can inhibit the perception of water-soluble taste compounds reacting with taste receptors on the tongue (Forss, 1969). Foods that contain emulsions of oil and water that form globules (oil surrounding water-based substance), and this can affect the perception of flavor compounds as well (Forss, 1969).

Proteins interact with flavor molecules and affect the retention and release of the flavor compounds based on the protein composition and product structure. Flavors can react with proteins in either reversible or irreversible bonds. Reversible reactions are physicochemical that consist of hydrogen bonds, hydrophobic interactions, and ionic bonds, while irreversible bonding consists of covalent bonds (amide and ester formation) and the condensation of aldehydes with amino and sulfhydryl groups (Kühn et al., 2006). Most flavors show hydrophobic, reversible reactions to proteins, but overall, there is no universal mechanism for flavors binding to proteins (Kühn et al., 2006).

Carbohydrates act similar to proteins in that their effect on flavor retention and release depends on the conditions and on the actual flavor molecules. Mono- and di-saccharides can affect flavor perception by binding to water molecules, causing unequal distribution of flavor and concentration of flavor in the remaining available water. Cyclic oligosaccharides and polysaccharides form complexes that encapsulate flavor compounds, making them good flavor carriers and retainers (Ammari and Schroen, 2018). The retention and release of flavor compounds in carbohydrates are mainly influenced by molecular weight, chemical groups, and polarity (Goubet et al., 1998). Aroma compounds with high molecular weights exhibited high retention of flavor when entrapped by a carbohydrate due to their inability to diffuse through the matrix during drying (Reineccius, 1988). Low molecular weight compounds are smaller, and can

diffuse out of the carbohydrate matrix more easily. It is hypothesized that smaller aroma compounds do not provide sufficient binding to the carbohydrate cavity, thus having weaker strength binding to the wall. Multiple studies show that alcohols have a high retention of flavor in carbohydrates compared to other chemical groups (Le Thanh et al., 1992). This could be due to the polarity of the chemical groups in the aroma compounds, as polar chemical groups are known to be less retained by carbohydrates. Similar to how water diffuses out of carbohydrates during drying, polar aroma compounds that are soluble in water can bind with water and diffuse out of carbohydrate matrices and release (Rosenberg et al., 1990).

Sweet Taste Perception

The human mouth consists of receptors to distinguish taste, temperature and touch sensations in food. Chemical substances interact with nerve cell bundles in taste buds that create signals to the cranial nerves and central nervous system. Foods consisting of sugars (glucose, sucrose, maltose) and artificial sweeteners (saccharin, aspartame) are responsible for the perceived sweetness sensation on the tongue.

The three main types of taste buds located throughout the tongue are fungiform, folate, and circumvallate and are connected to nerve fibers that form neural pathways to the cranial nerves and brain (Roper and Chaudhari, 2017; AlJulaih and Lasrado, 2021; Gibbons and Sadiq, 2021). Taste receptor cells (TRC) are clusters on taste buds of the tongue that are activated by specific solvated chemicals to signal and trigger action potentials to transmit information via sensory afferent fibers from the peripheral taste buds to the cranial nerves (Lee and Owyang, 2017; AlJulaih and Lasrado, 2021).

Of the four subtypes of TRC's that have been identified, Type II cells express G-protein coupled receptors (GCPR) that detect sweet taste presence in food (Lee and Owyang, 2017).

GPCRs are transmembrane receptors that mediate transduction pathways in subsets of receptor cells and detects extracellular signals to transmit information via neurotransmitter signals to the cranial nerve and the cortical taste centers to analyze information recognized by TRC's (Adler et al., 2000; Gravina et al., 2013). Two classes of GPCRs are responsible for forming heterodimers to detect sweet taste presence, Type 1 taste receptor family members 2 (T1R2) and member 3 (T1R3) (Nelson et al., 2001; Chaudhari and Roper, 2010; Sanematsu et al., 2014). Compounds that elicit a sweet taste bind and activate to the T1R2 and T1R3 receptors, which consist of three principal domains: an extra cellular venus-flytrap (VFT) domain at the N terminus, a seven transmembrane-spanning domain at the C terminus, and a cystein-rich linker that joins them (Fernstrom et al., 2012). Not all sweet taste compounds interact with all three domains. Natural and artificial sweeteners such as sucrose, glucose and sucralose will bind and activate to the T1R2 and T1R3 receptors, while dipeptide sweeteners such as aspartame and neotame only bind to the T1R2 VFT domain, and sweet proteins like thaumatin and monellin can interact with both subunits and the cysteine-rick liners (Fernstrom et al., 2012). These sweetener-binding events lead to receptor interaction and activation that causes neurotransmitters to send signals for sweet taste information from the taste buds to the brain via the cranial and central nervous system where olfactory input and past experiences determine a response to the food (Fernstrom et al., 2012; Gravina et al., 2013).

Temporal Dominance of Sensations

Conventional sensory evaluation methods require panelists to make single-point measurements on sensations of foods that are reported as the maximum intensity or averaged intensity (Cliff and Heymann, 1993; Lawless and Heymann, 2010b). This method can miss important information regarding product sensations if two products have similar sensation

intensities but differ in the time to reach their peak intensity or duration (Lawless and Heymann, 2010b). Piggott (1994) and Dijksterhuis (1996) noted the process involving food consumption, like mastication, salivation, tongue movement, and swallowing, can modify the intensities and quality of aroma, taste, flavor and texture over time and can change the intensities throughout consumption (Di Monaco et al., 2014).

Temporal methods like Time-Intensity (TI) and Temporal Dominance of Sensations (TDS) are extensions of sensory scaling methods that measure the changes in perceived sensations over time (Ott et al., 1991; Cliff and Heymann, 1993; Lawless and Heymann, 2010b). Time-intensity has been utilized to measure temporal properties of sensory characteristics in model systems and commercial products (Leach and Noble, 1986; Ott et al., 1991; Cliff and Heymann, 1993; Duizer et al., 1993; Bonnans and Noble, 1995; Ishikawa and Noble, 1995; Dijksterhuis, 1996; King and Duineveld, 1999). Temporal methods have been commonly used to investigate the sensory characteristics of sweeteners due to the delayed onset of bitter and metallic tastes (Larson-Powers and Pangborn, 1978; Shamil et al., 1988; Ott et al., 1991; Cliff and Heymann, 1993; Ketelsen et al., 1993). Ott et al. (1991) investigated the sweet and bitter taste characteristics of aspartame, acesulfame K and alitame at equisweet concentrations to sucrose in water using time-intensity. Larson-Powers and Pangborn (1978) used time-intensity to measure the sensory characteristics of sweeteners in flavored drinks and gelatin.

Time-intensity requires panelists to judge the intensity of one attribute at a time in a sample, making this process time consuming if there are multiple samples and attributes to analyze. To reduce time and eliminate the need for extensive training on intensity scales, Temporal Dominance of Sensations requires panelists to select the most dominant attribute or taste at a particular time point from a set of attributes (Pineau et al., 2009). Developed in 1999 by

Pineau et al. (2003), TDS requires panelists to select a new dominant attribute that captures their attention, the most striking perception, or the new sensations that pops up at a given time but may not necessarily be the most intense sensation (Pineau et al., 2009; Di Monaco et al., 2014). This method does not require lengthy training of judges and several attributes can be measured simultaneously in TDS compared to TI, but judges must be highly motivated as TDS requires continuously selecting the dominant attribute (Pineau et al., 2003; Le Révérend et al., 2008; Ng et al., 2012). Many studies have used TDS to examine temporal sensory characteristics of food products (Labbe et al., 2009; Meillon et al., 2009; Albert et al., 2012; Ng et al., 2012; Bruzzone et al., 2013). When an attribute is selected as most dominant, the duration of time is recorded and used to calculate a dominance rate that can be graphed to show the dominance of the attribute over time for the entire panel. The dominance rate can be viewed as a reflection of consensus among judges, and a measurement of panel performance, thus the higher the dominance rate of the attribute, the better agreement among judges (Pineau et al., 2009; Albert et al., 2012). Dominance rates of attributes in a single sample produce TDS curves that are plotted against similar samples for comparison when the dominance rate is considered significantly different from zero (Meillon et al., 2009; Pineau et al., 2009; Albert et al., 2012; Bruzzone et al., 2013).

Bemfeito et al. (2016) used TDS to characterize textural and flavor differences in Minas artisanal cheeses. The TDS results highlighted textural differences such as creamy, and soft or hard and firm sensations, while other cheeses had high dominance rates of crumbly texture (Bemfeito et al., 2016). Regarding flavor, most of the artisanal cheeses displayed high dominance rates of salty and bitter tastes, while some cheeses were characterized by either a sour taste or buttery and rancid attributes, which indicated a lack of product standardization (Bemfeito et al., 2016). Morais et al. (2014) used TDS to evaluate chocolate dairy desserts containing

prebiotic and sucrose substitutes. The study found similarities in the dominance rates between the sucralose and aspartame samples to the sucrose control dairy dessert sample based on sweetness, bitterness, milk chocolate flavor, bittersweet chocolate flavor, milk powder, cream and off-flavor attributes (Morais et al., 2014). The use of prebiotics also provided temporal profiles curves that were close control chocolate dairy dessert sample with no prebiotics (Morais et al., 2014). Bruzzone et al. (2013) used TDS to characterize and evaluate the textural dynamics of yogurt influenced by varying levels of fat, modified starch, and gelatin concentrations. Thomas et al. (2015) used TDS to characterize the dominant attributes in gouda cheeses at different ages and fat contents. This study found significant differences in preference for melting, fatty and tender texture attributes, and indicated which attributes should have a longer dominance duration in order to please specific consumers targets (Thomas et al., 2015). Esmerino et al. (2017) used TDS to evaluate fermented dairy strawberry yogurt and milk beverages. These results showed that most of the samples had bitter and sour tastes as a dominant attribute at some point in time due to lactic acid bacteria found in the fermented dairy products (Esmerino et al., 2017). Temporal Dominance of Sensations is an effective sensory profiling method that provides supplementary information on the evolution of sensory characteristics during food consumption that are not measurable by TI or by conventional sensory analysis.

Objectives

With milk consumption decreasing among children and adolescents, it is more imperative to find a healthy alternative to sweetened chocolate milk that contains no added sugar and is acceptable with parents and children. The objective of this study was to evaluate consumer perception of school lunch chocolate milk with lactose hydrolysis sweetened with nonnutritive sweeteners using descriptive analysis techniques and consumer evaluation.

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CHAPTER 2: THE ROLE OF SUGAR REDUCTION ON FLAVOR AND ACCEPTANCE IN SCHOOL LUNCH CHOCOLATE MILK.

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INTERPRETIVE SUMMARY

Sucrose replacement in school lunch chocolate milk with nutritive and nonnutritive sweeteners with and without lactose hydrolysis of milk was investigated. The application of blends of nonnutritive sweeteners in combination with lactose hydrolysis proved effective at mitigation of sensory differences from sucrose sweetened chocolate milk while providing a lactose free no added sugar chocolate milk that was acceptable to adults and children.

ABSTRACT

Our objective was to investigate sucrose replacement in school lunch chocolate milk with nutritive and nonnutritive sweeteners with and without lactose hydrolysis of milk on adult and child acceptance. In experiment 1, chocolate milk was formulated with nutritive and nonnutritive sweeteners (allulose, mixed sugar syrup, stevia, monk fruit, or sucralose) to the iso-sweet intensity of a current school lunch chocolate milk formulation with 3.69% sucrose (w/v). In experiment 2, lactose-hydrolyzed chocolate milk was formulated with nutritive and nonnutritive sweeteners (allulose, mixed sugar syrup, stevia, monk fruit) to the iso-sweet intensity of 3.69% sucrose (w/v). In experiment 3, lactose-hydrolyzed chocolate milk was formulated with sweetener blends of allulose, monk fruit and stevia to the iso-sweet intensity 3.69% sucrose (w/v). Iso-sweetness for all experiments was determined using magnitude estimation scaling and confirmed by 2-alternative forced choice paired comparison tests. Sensory properties of chocolate milks in each experiment were documented by descriptive analysis (DA) and temporal dominance of sensations (TDS). Consumer acceptance of chocolate milks was evaluated by young adult consumers for all experiments, and by child consumers for experiment 3. Sweeteners in unhydrolyzed chocolate milk (experiment 1) were differentiated ($p<0.05$) from the sucrose control chocolate milk due to bitter and metallic tastes inherent in the sweeteners. Liking scores for chocolate milks sweetened with allulose, mixed sugar syrup and sucralose were not different ($p>0.05$) from the sucrose control chocolate milk while liking scores for chocolate milks sweetened with stevia and monk fruit were lower than the sucrose control ($p<0.05$). The application of lactose-hydrolysis to milk (experiment 2) increased sweetness and reduced the amounts of each sweetener needed for isosweet taste by approximately one-third compared to unhydrolyzed chocolate milk. Consumer acceptance of lactose hydrolyzed chocolate milks were

not different ($p>0.05$). The application of sweetener blends in lactose hydrolyzed chocolate milk resulted in minimal differences by DA and TDS, and no differences ($p>0.05$) in overall liking to the sucrose control chocolate milk among adult and child consumers. The application of blends of nonnutritive sweeteners in combination with lactose hydrolysis proved effective at mitigation of sensory differences from sucrose sweetened chocolate milk while providing a lactose-free no added sugar chocolate milk. The findings from this study can be useful for milk manufacturers in accomplishing the objective of a school lunch milk that is appealing to parents and children.

Key Words: chocolate milk, sucrose replacement, child acceptance

Introduction

Milk and dairy products provide essential nutrients for the growth and development of young children. Children who consume milk are more likely to have a higher dietary calcium intake, larger skeletal structure, and higher bone mass compared to children who do not consume milk (Black et al., 2002; Murphy et al., 2008; Rafferty and Heaney, 2008). Calcium consumption via intake of dairy rich foods like yogurt, milk and cheese is important because humans continue to grow even after childhood and dietary calcium consumption can prevent the occurrence of osteoporosis and bone fracture (Chan et al., 1995; Bonjour et al., 1997; Huth et al., 2006). The 2010 US Dietary Guidelines for Americans recommends that U.S. children and young adults consume 3 cups of milk or milk products per day to meet the recommended calcium intake (1,000 mg for 4 to 8 year old, 1,300 mg for 9 to 13 year olds) (Institute of Medicine (US) Committee to Review Dietary Reference Intakes for Vitamin D and Calcium, 2011).

To ensure children are meeting the nutritional recommendations by the U.S. Dietary Guidelines, schools are required to serve milk as part of the federal school meal programs such as the National School Lunch Program (NSLP) and the School Breakfast Program (SBP). These programs aim to provide nutritious and well-balanced meals to children in low-income households (Code of Federal Regulations, 2019). Schools that participate in the NSLP and SBP receive cash subsidies from the programs for every meal served that follows the most recent Dietary Guidelines for Americans (Sipple et al., 2020). In 2010, the U.S. government initiated the Healthy, Hungry-Free Kids Act that established a nutritional standard for meal and beverages provided through school meal programs. The goal of this new Act was to improve the health and nutrition of children to prevent obesity and other health-related issues by offering more fruits and vegetables, more whole grain products, and reducing the sugar and calorie content of foods

served in schools (Kenney et al., 2020). This Act forced schools to remove 2% fat unflavored and flavored milk from school menus if they wanted to continue receiving cash subsidies. The decline of milk consumption and participation in school meal programs caused the USDA to initiate an interim rule called the "Flexibilities for Milk, Whole Grains, and Sodium Requirement" that allowed flexibilities to the Child Nutrition Program menus, one of which included re-introducing 1% fat flavored milk in schools.

Studies have shown that children view taste as an important attribute in the milk they consume, and prefer flavored milk over unflavored (Johnson et al., 2002; De Pelsmaeker et al., 2013; Henry et al., 2015). When flavored milk was removed from school menus, total milk consumption decreased by 35% (MilkPEP, 2017). Despite flavored milk's popularity in schools, some parents remained concerned about children consuming excess amounts of sugar. Li et al. (2014) reported that parents were most concerned about the type of sweetener and fat content in children's milk, and preferred that children consume flavored milk with reduced sugar instead of no added sugar or regular sugar content. Despite the concerns for sugar content in milk, parents preferred that children consume any milk over juice or soda (Fayet-Moore, 2016; Racette et al., 2023). Racette et al. (2023) reported that parents demonstrated a low awareness of milk nutrition, but valued flavored milk as a vehicle for nutrition for their children, regardless of the added sugar content. Providing reduced-sugar flavored milk allows access to essential nutrients that children need for growth, and prevents child consumption of unhealthy sugar-sweetened beverages like soda and juice.

To meet the demands of parents and children, sugar-reduction methods such as nonnutritive sweeteners and lactose hydrolysis have been researched on dairy products. Previous studies have shown that sugar content in various foods can be reduced by 25% to 30% while

maintaining consumer acceptance (Li et al., 2015a; McCain et al., 2018). Studies have also reported that partial replacement of sucrose with alternative sweeteners such as stevia or monk fruit in dairy products can produce a similar sensory profile and consumer acceptance among adults and children (Li et al., 2015a; Velázquez et al., 2021; Chadha et al., 2022; Mora et al., 2022). Li et al. (2015a) reported that chocolate milk partially sweetened with stevia or monk fruit at a 75/25 ratio (sucrose/alternative sweetener) had a similar sensory profile to sucrose-sweetened chocolate milk. Bordi et al. (2016) and Verruma-Bernardi et al. (2015) also reported that chocolate milk sweetened with mixture of sucrose and stevia was more acceptable than stevia sweetened milk among adult and child consumers. Racette et al. (2023) reported segmentation among parents on the use of sugar alternatives in chocolate milk, with some parents highly favoring the use of these ingredients, while others did not.

Previous studies have also investigated hydrolysis of native lactose with or without added (additional) lactose as a sugar-reduction method in milk. Li et al. (2015b) reported that hydrolysis of >7.5% of added lactose followed by lactose hydrolysis was needed to achieve the sweetness level of 150 n*M* sucrose-sweetened chocolate. The lactose hydrolyzed chocolate milk was not different in liking from a sucrose control chocolate milk, but contained additional calories to the milk due to the added lactose (Li et al., 2015b). Lactose-free milk is traditionally manufactured by enzymatic hydrolysis of lactose and results in a lactose free milk that is sweeter and less viscous than traditional milk (Adhikari et al., 2010; Rizzo et al., 2020).

To formulate sucrose replacement in a food, the sweetness equivalence (isosweetness) or the amount of alternative sweetener that produces the same sweetness intensity, must be determined in the target food matrix. Magnitude estimation scaling (MES) followed by descriptive analysis and 2-alternative forced choice (2-AFC) testing is a standard method to

determine sweet taste equivalency and used in past studies (Cardello et al., 1999; Li et al., 2015a; Parker et al., 2018). Alternative sweeteners like stevia, monk fruit, and sucralose are well established alternative zero-calorie sweeteners that are useful to replace sucrose because of their sweet taste intensity and lack of calories but can have different sensory profiles due to off-flavors like bitter and metallic tastes (Kim and Kinghorn, 2002; Morais et al., 2014; Tan et al., 2019). Allulose is a C-3 epimer of fructose that naturally occurs in wheat and processed sugar cane and has approximately 70% of the sweetness of sucrose (Xia et al., 2021; Han et al., 2016). Allulose is considered a very low-calorie sweetener at 0.4 calories per gram and has the potential to appeal to consumers due to its similar flavor profile to sucrose (Jürkenbeck et al., 2022).

When replacing sucrose in a product, the temporal sensory profile of nonnutritive sweeteners is important to understand because each sweetener has a specific sensory profile due to its sweet taste intensity and additional flavors and tastes. Temporal methods have also been applied to examine sensory profiles of dairy products including cheese, milk, protein beverages and yogurt (Morais et al., 2014; Esmerino et al., 2017; Thomas et al., 2017; Keefer et al., 2020; Harwood and Drake, 2021; Chadha et al., 2022). The purpose of this study was to investigate sucrose replacement in school lunch chocolate milk with nutritive and nonnutritive sweeteners with and without lactose hydrolysis of milk on adult and child acceptance. Allulose, mixed sugar syrup (fructose, allulose, glucose), monk fruit, stevia and sucralose were selected as sweeteners for this study. Mixed sugar syrup was included in this study as a natural reduced-sugar sweetener blend, as it contains 2 calories per gram.

Materials and Methods

Experimental Overview

Chocolate milk (experiment 1) and lactose hydrolyzed chocolate milks (experiments 2 and 3) were formulated with nutritive and nonnutritive sweeteners to the iso-sweet intensity of a current school lunch chocolate milk formulation with sucrose **(**Figure 1). Iso-sweetness was confirmed in each chocolate milk using magnitude estimation scaling (MES) and 2-alternative forced choice (2-AFC) paired comparison tests with consumers. The chocolate milks were profiled using descriptive analysis (DA) and temporal dominance of sensations (TDS) with trained panelists. Chocolate milks were evaluated for acceptance by adult consumers (experiments 1, 2 and 3) and by child consumers (experiment 3) alongside the current school lunch chocolate milk with added sucrose. All sensory testing was performed in compliance with the North Carolina State University Institutional Review Board for Human Subjects regulations (NCSU IRB).

Experiment I: Sugar Reduction of School Lunch Chocolate Milk

Chocolate Milk Manufacture. School lunch chocolate milk (chocolate milk) was manufactured in the pilot plant at North Carolina State University (Raleigh, NC) using a formula consistent with current school lunch guidelines (Keefer et al., 2022). Five hundred liters of 1% milkfat raw milk (somatic cell count (SCC) < 100,000) was obtained from the North Carolina State University Dairy Enterprise System. Milkfat content of raw milk was confirmed by a Fourier Transform Mid-Infrared (FTIR) spectrophotometer (Lactoscope FTA, Delta instruments, Drachten, Netherlands).The FTIR was calibrated using modified milk samples as described by (Kaylegian et al., 2006). Raw chocolate milk base (without added sugar) with cocoa powder (0.60% w/v) (cocoa powder blend #5, Tate and Lyle, Decatur, IL, USA) was pasteurized in a Microthermics ENVH unit (Microthermics, Raleigh, NC, USA) at 78°C for 26 seconds, then

immediately fed into a 2-stage homogenizer (model NS2006H, GEA Niro Soavi, Parma, Italy) at 13.8 Mpa (Stage 1=10.3 MPa; Stage 2=3.4 Mpa). The unsweetened pasteurized chocolate milk base was then filled into 40-liter milk cans, cooled to 4°C in an ice bath and stored at 4°C. Legal pasteurization of milk was confirmed by alkaline phosphatase testing (ZymoSnap ALP, Hygiena, CA, USA). Samples of the chocolate milk base were taken and plated in triplicate for aerobic plate count (**APC**) and coliform count (**CC**) and incubated at 35°C for 24 and 48 h, respectively, using Petrifilm™ plates (Aerobic Count Plates and Coliform Count Plates, 3M™, St. Paul, MN).

Nutritive and nonnutritive sweeteners were added to the pasteurized unsweetened chocolate milk base for all experiments. Sweeteners were added to chocolate milk at 4°C and stirred thoroughly. The next day, milks were mixed thoroughly again, and sensory tests were completed within 3 days. Granulated cane sugar was purchased from a local grocery store. Allulose syrup (Dolcia Prima® allulose, 75%), stevia (TASTEVA™, 95% steviol glycosides), mixed sugar syrup (C2-331 Mixed sugar syrup (fructose, allulose, glucose)), and monk fruit (PUREFRUIT™ Select, 48% mogroside V) were provided by Tate & Lyle (Decatur, IL, USA). Sucralose was purchased from Bulk Supplements (Henderson, NV, USA). Total solids and fat were measured in duplicate for each chocolate milk.

Power Function Curves of Sweeteners in Chocolate Milk. Power function curves for allulose, mixed sugar syrup, monk fruit, stevia, and sucralose were generated in water and chocolate milk with trained panelists to determine sweet taste equivalence to 3.69% sucrose in 1% fat chocolate milk (current sucrose added to school lunch chocolate milk) and 5.75% sucrose in deionized water (sweet taste intensity of 3.69% sucrose in 1% chocolate milk). Each panelist (n=8, 4 females, 4 males, ages 21-30 y) had a minimum of 30 h of previous experience with descriptive sensory analysis and an additional 12 h of training with Magnitude Estimation Scaling (MES).

Panelists were confirmed to demonstrate replicable ratio scaling with a range of sweetened solutions prior to data collection (Li et al., 2014; Wagoner et al., 2018; Keefer et al., 2020).

For the water portion of MES, a reference solution of 5.75% sucrose represented a reference intensity of 100, and scale anchors of 2.87% and 11.5% w/w sucrose represented intensities of 50 and 150 respectively. For chocolate milk, a reference solution of 3.69% w/w sucrose in chocolate milk represented the reference intensity of 100, while 1.85% and 7.38% w/w sucrose in chocolate milk represented intensities of 50 and 150 respectively. Panelists were instructed to refer to these references as they tasted each test treatment. Seven sweetener concentrations of each treatment were formulated using a geometric scaling factor of 1.4 (Parker et al., 2018). Initial concentrations of each sweetener were determined based on literature values and preliminary descriptive analysis (DA). Sweeteners were added to water and chocolate milk base separately at a step factor of 1.4 to create 6 subsequent solutions (three below and three above the theoretical iso-sweet concentration) for a total of 7 samples for each matrix (water and chocolate milk).

Power functions for each sweetener were generated first in water, then in the chocolate milk base to determine iso-sweet concentration to the current school lunch formulation with sucrose. No more than seven samples were presented during any session. Panelists were presented with samples at 4°C coded with 3-digit numbers in a random order and asked to scale the maximum perceived sweetness to the reference solution. A 4-minute rest was enforced between each sample. Panelists were instructed to rinse their mouth with water and take a bite of an unsalted cracker between samples. Each sweetener was evaluated in each matrix in triplicate and data was collected using Compusense Cloud (Compusense, Guelph, Canada). The iso-sweet concentration of each sweetener in water and chocolate milk was then calculated using a power

function curves (Cardello et al., 1999). Descriptive analysis was performed to confirm and adjust iso-sweetness concentrations for each sweetener in each matrix. Subsequently, the iso-sweet concentration of each sweetener in water and in chocolate milk was then confirmed using a directional paired comparison test with consumers (n=40) (2-Alternative Forced Choice, 2-AFC) (ASTM, 1991).

Descriptive Analysis. Eight trained descriptive analysis panelists (4 females, 4 males, ages 21-55 y) each with > 150 h of descriptive analysis experience with dairy products including chocolate milks, evaluated the chocolate milks at iso-sweet concentration in duplicate using an established sensory language (Table 1, Li et al., 2014) and a 0 to 15-point intensity scale consistent with the Spectrum descriptive analysis method (Meilgaard et al., 2016). Milks were presented in lidded 60-ml soufflé cups with 3-digit codes and were evaluated at 10C. Data was collected using a secure server on the NCSU secure network.

Temporal Dominance of Sensations. Perception of temporally dominant attributes in iso-sweet chocolate milks was determined using Temporal Dominance of Sensations (TDS) with a trained panel (n=8, 4 females, 4 males, ages 23-54 y) (Pineau et al., 2009). Trained panelists (described previously) participated in an additional 16 h of experience with TDS and chocolate milks prior to data collection. Panelists were provided with 20 ml samples presented in 60 ml soufflé cups with 3-digit codes using a balanced presentation order. Panelists were instructed to put the entire sample in their mouth, start the timer, and to select the most dominant attribute. The dominant attribute was defined as the attribute that drew the panelist's attention (Labbe et al., 2009; Pineau et al., 2009; Lawless and Heymann, 2010b; Di Monaco et al., 2014). The attributes used in temporal profiling consisted of taste, flavor, and oral-tactile sensation attributes, including sweet taste, bitter taste, metallic, sweet aromatic flavor, cardboard flavor, and astringency. Panelists

could also select “none” as an option. Panelists were prompted to expectorate the sample at 15 s and to evaluate the sample over the course of 192 s (Parker et al., 2017). Each sample was evaluated by each panelist in triplicate in separate sessions. Data was collected using Compusense Cloud (Compusense, Guelph, Canada).

Adult Consumer Acceptance. Six chocolate milks (allulose, mixed sugar syrup, monk fruit, stevia, sucralose at isosweet concentration and the 3.69% sucrose control) were evaluated by adult chocolate milk consumers. Chocolate milks (1% milkfat) were manufactured in the North Carolina State University pilot plant as described previously and were stored and served at 4°C (Table 1). Consumers (n =124) were recruited from the consumer database of more than 10,000 people maintained by the NCSU Sensory Service Center using an online screener (Compusense, Guelph, Canada). To participate, panelists were required to be self-reported chocolate milk consumers (consumed chocolate milk within the past 3 months) and consumers of 1% fat milk.

Milks were presented monadically in a Williams design to prevent order of presentation effects. Consumers had a 3-min enforced rest with bottled spring water and unsalted cracker between samples. Each chocolate milk (60 ml) was poured into a 4-ounce clear plastic tumbler labeled with 3-digit codes. Consumers were asked to evaluate the liking of appearance, color, aroma, flavor, sweetness, cocoa flavor, mouthfeel/thickness, and aftertaste of each chocolate milk. Liking questions were evaluated on a 9-point hedonic scale, where 1=dislike extremely and 9=like extremely. Just about right (JAR) questions were also asked for color, aroma, flavor, sweetness, cocoa flavor, mouthfeel/thickness, and aftertaste (1 or 2=too little, 3=just about right, and 4 or 5=too much). Aftertaste liking and JAR questions were only asked to consumers who indicated they detected an aftertaste when tasting the sample of chocolate milk. Consumers also rated the aftertaste pleasantness of each sample, where 1 or 2=unpleasant, 3=neither pleasant nor

unpleasant, and 4 or 5=pleasant. At the end of each sample, consumers indicated if the chocolate milk sample met their expectations, where 1 or 2=did not meet expectations, 3=meets expectations, and 4 or 5=exceeds expectations. Consumers also indicated their purchase intent of each chocolate milk where 1 or 2=will not buy, 3=might or might not buy, and 4 or 5=will buy. After tasting all chocolate milks, consumers answered demographic questions. After completion of the test, participants were compensated with a gift card to a local shopping store.

Experiment II: Sweetener Reduction using Lactose Hydrolysis

Chocolate Milk Manufacture. Chocolate milk manufacture proceeded as previously described with the following modifications. Lactase enzyme (Saphera 2600, Novoymes, Franklinton, NC, USA) was incorporated into the raw 1% milk to hydrolyze existing lactose into glucose and galactose. The enzyme was added to raw 1% milk at a usage rate of 1 gram of enzyme per liter of raw milk as recommended by the supplier then held at 4°C for 24 h or until milk reached 97% hydrolysis of lactose. Lactose hydrolysis was confirmed by cryoscope (Advanced Instruments, Model 4250, Norwood, MA, USA) by measuring the linear relationship between freezing point depression and enzymatic activity (Baer et al., 1980; Chen et al., 1981). Following lactose hydrolysis, processing proceeded as previously described.

Power Function of Sweetness in Chocolate Milk. A power function curve for sucrose in lactose hydrolyzed milk was generated to determine iso-sweet concentration to the current chocolate milk (3.69% sucrose in unhydrolyzed chocolate milk). Reference solutions of 3.69% w/w sucrose in unhydrolyzed chocolate milk represented the reference intensity of 100, while 1.85% and 7.38% w/w sucrose in unhydrolyzed chocolate milk represented intensities of 50 and 150 respectively. Panelists (described previously) were instructed to follow the same procedure as described for experiment 1 for MES. Descriptive analysis followed by a directional paired

comparison test (n=40) (2-Alternative Forced Choice, 2-AFC) were then used to confirm isosweet concentrations (ASTM, 1991). 2.53% sucrose in lactose hydrolyzed chocolate milk was determined to be the iso-sweet concentration to 3.69% sucrose in unhydrolyzed chocolate milk and was used as a reference for power function curves with the alternative sweeteners in lactose hydrolyzed chocolate milk.

Then, power function curves for allulose, mixed sugar syrup, stevia, and monk fruit were generated to determine iso-sweet concentration of sweeteners in lactose hydrolyzed chocolate milk to 2.53% sucrose in lactose hydrolyzed chocolate milk. A reference solution of 2.53% sucrose in lactose hydrolyzed chocolate milk represented a reference intensity of 100 (2.53% sucrose in lactose hydrolyzed chocolate milk is the equivalent sweetness to 3.69% sucrose in unhydrolyzed chocolate milk and scale anchors of 1.3% and 5.1% w/w sucrose represented intensities of 50 and 150 respectively. Panelists (described previously) were instructed to follow the same procedure as described for experiment 1 for MES. Descriptive analysis followed by a directional paired comparison test (n=40) (2-Alternative Forced Choice, 2-AFC) were then used to confirm isosweet concentrations (ASTM, 1991). Iso-sweetness of each sweetened lactose hydrolyzed milk to the 3.69% sucrose in unhydrolyzed chocolate milk (sucrose control) were also confirmed subsequently by paired comparison tests as described previously.

Descriptive Analysis and Temporal Dominance of Sensations. Descriptive analysis and TDS of sweeteners in lactose-hydrolyzed chocolate milk (Table 2) followed the same procedure as described in experiment 1.

Adult Consumer Acceptance. Lactose hydrolyzed chocolate milks (n=5) (allulose, mixed sugar syrup, monk fruit, stevia, 2.53% sucrose) and the 3.69% unhydrolyzed chocolate milk (sucrose control) were evaluated by adult chocolate milk consumers (Table 2). Lactose hydrolyzed

chocolate milks and unhydrolyzed chocolate milk (sucrose control) were manufactured as previously described. Consumers (n=160) were recruited were recruited as described previously and the same ballot was used as in experiment 1.

Experiment III: Sweetener Reduction using Lactose Hydrolysis and Sweetener blends

Lactose Hydrolyzed Chocolate Milk Manufacture. Lactose hydrolyzed school lunch chocolate milk base was produced using the procedures described previously.

Power Function of Sweetness in Lactose Hydrolyzed Chocolate Milk with Sweetener Blends. The sweetness equivalence concentration of each individual sweetener was used to formulate sweetener blends. Iso-sweetness of each blend to the 3.69% sucrose in unhydrolyzed chocolate milk (sucrose control) were confirmed by descriptive analysis and paired comparison tests as described previously. Sweetener blends (Table 3) containing stevia, monk fruit and allulose were generated and were selected based on price per serving and novelty of the sweetener blends compared to previous research studies (Li et al., 2015a; Parker et al., 2018).

Descriptive Analysis and Temporal Dominance of Sensations. Descriptive analysis and TDS for sweetener blends in lactose hydrolyzed chocolate milk were conducted as previously described (Table 3).

Adult Consumer Acceptance. Four sweetener blends in lactose hydrolyzed chocolate milk (Table 3), the sucrose LH chocolate milk and the sucrose control chocolate milk were evaluated by adult chocolate milk consumers (n=131). Milks were manufactured as previously described and procedures for the adult consumer test also proceeded as previously described.

Child Consumer Acceptance. The sucrose lactose hydrolyzed chocolate milk, the sucrose control chocolate milk, and three sweetener blend formulations of lactose hydrolyzed chocolate milks were selected for evaluation for acceptance by child chocolate milk consumers (Table 4). The

five chocolate milk formulations were manufactured as previously described. Children aged 8-13 y that consumed milk at school at least 2-3 times a month and consumed fat free or low-fat flavored milk were recruited with parental consent from the previously described consumer database (n=128). Children were recruited based on parental self-reported frequency of milk consumption and were required to have no food allergies, intolerance or dietary restrictions.

Child consumer testing was performed in January 2023 and was modified for COVID-19 safety per university IRB restrictions that remained in place for minors at that time (Racette and Drake, 2022; Schiano et al., 2022). Parents/legal guardians of participating children were instructed to pick up coded chocolate milk samples in a thermal bag (ULINE) with a bag of unsalted saltines and a commercial bottle (2) of spring water. After providing written parental/legal guardian consent at the time of sample pickup, parents/legal guardians were instructed to take the samples home and click on the hyperlink to complete a Zoom video call to obtain child consent. Verbal child consent was obtained from each child participating in the test during each of two virtual tasting sessions conducted via Zoom (version 5.13.10, Zoom Video Communications, Inc., CA, USA,). During each scheduled time, the children were asked if they would like to participate in the test before they began each tasting session. Children were required to consent to participate in the research for each day of testing (2 days). Once child consent was obtained, parents were given a login code for their child/children participant(s) and used a hyperlink to Compusense to initiate the taste test. Video oversight by the researchers was present throughout each test session to ensure protocol was followed and to answer any questions from the child.

Samples were presented monadically in a Williams Design with a partial presentation across two days, where children evaluated three chocolate milks on Day 1, then two on Day 2.

Children had an enforced 3-min rest with water and unsalted cracker rinse between samples. For child consumer testing, each chocolate milk (60 ml) was poured into a 240 mL opaque PET bottle (Consolidated Container Company, New Castle, PA, USA) under a laminar flow hood. Each chocolate milk was labeled with a 3-digit code for sample identification. Children were asked to evaluate appearance, aroma and overall liking of each chocolate milk. Liking questions were evaluated on a 7-point hedonic scale (Pagliarini et al., 2003; Laureati et al., 2015), where 1=dislike extremely and 7=like extremely. Just about right (JAR) questions were asked for cocoa flavor and sweetness (1 or 2=too little, 3=just about right, and 4 or 5=too much). Questions regarding purchase intent (1 or 2= would not want parent to buy, 3= might or might not want parent to buy, 4 or 5=want parent to buy) and how the chocolate milk samples compared to their current school lunch chocolate milk (1 or 2= I like it less, 3= I like it about the same, 4 or 5= I like it more) were also asked to the children after the liking and JAR questions. After completion of the test, parents of the child participants were compensated with a gift card to a local shopping store. Compusense Cloud (Compusense, Guelph, Canada) was used to collect the data.

Statistical Analysis

Iso-sweet concentrations were generated for each sweetener by graphing the dose-response power function from MES and calculating the sweetener concentration required for intensity equivalent to the reference 100 or 3.69% w/w sucrose in unhydrolyzed chocolate milk for power function curves in experiment 1 and 2. Statistical analysis of power function slopes and equivalent sweetener concentrations was performed using two-way ANOVA for matrix x sweetener interaction with Fisher's least significance difference test to determine means separation (alpha = 0.05) using XLSTAT (Version 2023.2.2, Addinsoft, New York, NY, USA). Statistical significance of 2-AFC paired comparison tests were determined based on the

minimum number of correct judgements for significance at alpha = 0.05 (Meilgaard et al., 2007). Temporal dominance of sensations data from TDS were presented as curves where attribute dominance rates were plotted over time using XLSTAT (Version 2023.2.2, Addinsoft, New York, NY, USA), and a smoothing function within R Studio software was used to construct the curves. Significance was calculated at 95% confidence. Comparisons between sweeteners and milks were made at each second of evaluation using 2-sided Fisher's exact test. Citation proportions and differences were graphed using XLSTAT (Version Addinsoft, New York, NY, USA). Analysis of variance (ANOVA) with mean separation using Fisher's least significant difference was performed on descriptive analysis data, consumer liking and intensity data. JAR data for consumer test results were reported in percentages and analyzed using penalty analysis; significant differences for JAR data were determined using chi-square. Liking data for children and non-JAR questions were analyzed using Kruskal-wallis nonparametric test. All statistical analyses were performed at the 95% confidence interval.

Results

Experiment I: Sugar Reduction of School Lunch Chocolate Milk

Power Function of Sweetness in Chocolate Milk. The sweet taste equivalence of each nutritive and nonnutritive sweetener (NNS) was determined and measured to confirm that the differences in chocolate milk liking were not due to a perceived difference in sweet taste but rather due to other sensory attributes specific to each sweetener. Power function curves display a linear relationship between intensity of the perceived stimulus and the concentration of the stimulus, and the exponent or slope of the power function denotes the rate of change in the perceived stimulus (i.e. sweetness) as a function of concentration (Cardello et al., 1999; Lawless and Heymann, 2010b; Wagoner et al., 2018). The dotted line at y=2 intercept indicates the scaled

sweetness intensity equivalent of the designated reference (3.69% sucrose chocolate milk) and the intersections of the individual sweetener power functions represent the concentration required for iso-sweetness with 3.69% sucrose chocolate milk (Figure 2). Sucralose was the most potent sweetener, followed by stevia, monk fruit, sucrose, mixed sugar syrup and allulose, and results for sweetness intensity are consistent with current literature (Table 5)(US Food and Drug Administration, 2018). The power function slopes for each sweetener were higher in water than in chocolate milk (Table 6) meaning that a lower addition of sweetener was required to initiate a change in sweet taste in water compared to a more complex matrix, chocolate milk. Studies have indicated that a smaller power function slope represents more potent sweetness intensity and thus would require less mass of sweetener to match the sweetness equivalence (Li et al., 2015a; Wagoner et al., 2018).

Descriptive Analysis. Trained panelists documented sensory differences other than sweet taste among the chocolate milks (Table 7). Monk fruit, stevia and sucralose sweetened chocolate milks had low but distinct metallic taste, and stevia and monk fruit sweetened chocolate milks were also characterized by low bitter taste, which is consistent with sensory profile results from (Jung et al., 2021). This is consistent with (Parker et al., 2018) and (Kim et al., 2015) that also found bitter and metallic tastes in products sweetened with nonnutritive sweeteners through descriptive analysis. In general, milks sweetened with allulose, sucralose, or mixed sugar syrup had the most similar sensory profiles to chocolate milks sweetened with sucrose.

Temporal Dominance of Sensations. Temporal dominance of sensations (TDS) was applied to chocolate milks with trained panelists to document sensory differences that might not be directly apparent by traditional descriptive analysis (Keefer et al., 2023). By TDS, the ideal goal is to have temporal dominance profiles of chocolate milk sweetened with alternative sweeteners be

identical to the sucrose control chocolate milk. Chocolate milks with allulose, mixed sugar syrup and sucralose chocolate milks had dominance profiles most similar to the sucrose control in chocolate milk due to their high initial sweet taste, followed by astringency (Figure 3). Chocolate milks with monk fruit or stevia were characterized by a dominant sweet taste, followed by bitter and metallic tastes, then astringency. The monk fruit chocolate milk was higher in metallic taste than the sucrose control chocolate milk. The temporal dominance curves of chocolate milk sweetened with monk fruit and stevia are consistent with descriptive analysis results and previous temporal studies that associated these sweeteners with bitter and metallic tastes in addition to sweetness (Cardello et al., 1999; Morais et al., 2014; Reyes et al., 2017; Chadha et al., 2022). All chocolate milks exhibited similar results in TDS of an initial sweet taste, followed by astringency. The results from TDS, along with descriptive analysis results, indicate that these nonnutritive sweeteners were similar to sucrose when applied at iso-sweet concentration to sucrose in chocolate milk, but sensory differences that may impact consumer acceptance still remained.

Adult Consumer Acceptance. Adult chocolate milk consumers (n=122, 19-64 y) were 31.1% male and 67.9% female and were mostly Caucasian (68.9%) and Asian/Pacific Islander (17.2%) who consumed fluid milk daily or weekly (> 89 %), consumed flavored milk weekly or 2-3 times a month (>63%) and preferred reduced fat or 2% milk (59%) (see Appendix J). There was no differences in appearance, color, aroma, and mouthfeel/thickness liking ($p>0.05$) among the chocolate milks (Table 8). Chocolate milk sweetened with mixed sugar syrup was not different in any liking attribute from the sucrose control chocolate milk ($p>0.05$). There were no differences documented among the chocolate milks for chocolate flavor liking ($p>0.05$) except for monk fruit which received a lower score ($p<0.05$). Chocolate milks sweetened with monk

fruit, stevia or sucralose received the lowest sweetness and aftertaste liking scores, which was consistent with descriptive analysis and TDS results that documented bitter and metallic tastes in these milks.

All chocolate milks were penalized for being too weak in flavor and chocolate flavor (Table 8). These results are expected since this school lunch chocolate milk was formulated to have a low amount of added sugar (Table 5), and a low amount of added sugar also means a low amount of added cocoa since cocoa has inherent bitter taste that is masked by the addition of sugar. Low cocoa flavor and sweet taste intensity in school lunch chocolate milk would be expected and were evident in the trained panel profile of the sucrose control milk (Table 7). It is also likely that increased sweet taste may also enhance perception of chocolate flavor intensity (Prawira and Barringer, 2009; Hopfer et al., 2022). Chocolate milk sweetened with monk fruit was penalized for being both not sweet enough and too sweet, which suggests that consumers did not like the sweet taste, but did not know how to score it, which was also observed by (Li et al., 2015a). Chocolate milks sweetened with stevia or monk fruit were penalized the most for having an aftertaste that was too strong, consistent with results from previous studies (Li et al., 2015a; Chadha et al., 2022).

Experiment II: Sweetener Reduction using Lactose Hydrolysis

Power Function of Sweetener and Sweetener Blends in Chocolate Milk. The addition of lactase increased the sweetness of the lactose-hydrolyzed (LH) chocolate milk base due to the presence of glucose and galactose, which have a higher relative sweetness compared to lactose (Tiefenbacher, 2017). As such, a lower concentration of sucrose and each additional sweetener was required to meet the iso-sweetness of 3.69% sucrose in chocolate milk compared to what was required for unhydrolyzed chocolate milk (Table 10). The relative sweetness of each

sweetener in LH chocolate milk (Table 10) was similar, as expected, to experiment 1 with no lactose hydrolysis (Table 5).

Descriptive Analysis. Sensory profiles of the LH chocolate milks were generally similar (Table 11). A low intensity of bitter and metallic tastes were observed in the LH chocolate milk sweetened with stevia, and metallic was also documented in LH chocolate milk sweetened with monk fruit, consistent with trained panel profiles from traditional chocolate milks (Table 7).

Temporal Dominance of Sensations. TDS curves from experiment 2 (Figure 4) were similar to experiment 1 (Figure 3) with a high initial sweet taste dominance followed by astringency. LH chocolate milk sweetened with stevia had a low frequency of bitter taste dominance, consistent with trained panel profiles. No temporal differences ($p>0.05$) were documented between the sucrose control chocolate milk and the nonnutritive sweeteners in lactose-hydrolyzed milk. The TDS curves for alternative sweeteners in LH chocolate milk were similar to the sucrose control (unhydrolyzed) chocolate milk, suggesting that the use of lactose hydrolysis and the need for the addition of less sweetener resulted in a sensory profile more similar to the sucrose chocolate milk control.

Adult Consumer Acceptance. Lactose hydrolyzed (LH) chocolate milks sweetened with allulose, stevia, monk fruit, mixed sugar syrup, or sucrose, along with the sucrose control chocolate milk with 3.69% sucrose were evaluated by adult chocolate milk consumers (Table 12). Demographics for Experiment 2 adult consumer test were similar to experiment 1 (see Appendix K). Unlike results from the consumer test with unhydrolyzed chocolate milks, there were no differences ($p>0.05$) among the sucrose control and any of the LH chocolate milks (Table 11). Milks were also not different for appearance, color, flavor, sweetness, and mouthfeel liking ($p>0.05$). Chocolate milks were penalized for too weak flavor and chocolate flavor. Light color

(too light), too thin and too strong aftertaste were also penalized in several of the milks (Table 11). Chocolate milks except for LH chocolate milk sweetened with monk fruit were not different in purchase intent scores ($p>0.05$).

Experiment III: Sweetener Reduction using Lactose Hydrolysis and Sweetener Blends

Power Function of Sweetener Blends in Chocolate Milk. Power function slopes for the iso-sweet concentration of allulose, stevia, monk fruit and sucrose in lactose hydrolyzed (LH) chocolate milk from experiment 2 (Table 6) were used to generate isosweet sweetener blends (LH blends 1-4, Table 3).

Descriptive Analysis. Once again, minor differences in sensory profiles of the LH chocolate milks with different sweetener blends were documented (Table 14). Bitter taste and metallic were not detected. Based on results from the current study, a combination of lactose hydrolysis and sweetener blends can minimize sweetener-associated off-flavors.

Temporal Dominance of Sensations. Results for temporal dominance of sensations (TDS) dominance curves from experiments 3 (Figure 5) were similar to experiments 1 and 2 (Figure 3, Figure 4) where chocolate milks were characterized by high initial sweet taste dominance, followed by astringency. No temporal differences ($p>0.05$) were documented between the sucrose control chocolate milk and sweetener blends in lactose-hydrolyzed chocolate milk. The TDS results for each sweetener blend in chocolate milk were similar to the sucrose control chocolate milk and LH sucrose chocolate milk, indicating that the use of sweetener blends alongside lactose hydrolysis can produce chocolate milk nearly identical in sensory profile to the sucrose control chocolate milk currently being served in schools.

Adult Consumer Acceptance. Demographics for experiment 3 adult consumer test were similar to experiment 1 and 2 (see Appendix L). Chocolate milks were at parity for appearance liking,

overall liking, aroma liking, flavor liking and chocolate flavor liking ($p>0.05$) (Table 13). The sucrose control chocolate milk scored significantly higher ($p<0.05$) for sweetness liking than chocolate milks with LH blends #1 and #3. All chocolate milks except the 3.69% sucrose control chocolate milk were penalized for too weak of aroma. All samples except LH blend #1 were penalized for too weak overall flavor. LH sucrose chocolate milk and LH blend #3 were penalized for being not sweet enough, while LH blends #1, #2 and #4 were penalized for being too sweet. All chocolate milks except LH blend #2 were penalized for being too weak in chocolate flavor. The LH sucrose chocolate milk and LH blends #2 and #3 were penalized for being too thin. The sucrose control chocolate milk and LH sucrose chocolate milks were the only chocolate milks not penalized for having too strong of an aftertaste.

Children Consumer Acceptance. Demographics for child consumers consisted of 53.9% males and 46.1% females between the ages of 8-9 years old (31.3%) and 10-13 years old (68.8%), who were either in public (89.1%) or private (10.9%) school, purchased milk at school daily or weekly (>84.4%), and consumes flavored low-fat (98.4%) or fat-free (52.3%) milk at school. Chocolate milks were at parity ($p>0.05$) for color liking, aroma liking, and overall liking. The chocolate milk samples were penalized for not enough chocolate flavor. The sucrose control and LH blend #2 chocolate milks were penalized for not sweet enough, while the LH blend #4 chocolate milk was penalized for being too sweet. The LH stevia chocolate milk was penalized for being too sweet and not sweet enough. These results are consistent with to the adult consumer tests in experiments 2 and 3 where no differences in overall liking scores were observed between LH stevia, LH blend #2 and LH blend #4 to the sucrose control chocolate milk (Table 12, Table 15). (Li et al., 2015a) found that chocolate milks sweetened with stevia: sucrose or monk fruit: sucrose blends were acceptable among children, which is consistent with the

results of this study that solely used nonnutritive sweetener blends in chocolate milk with no additional calories or added sugar.

Discussion

Results for relative sweetness of monk fruit, stevia, and sucralose in chocolate milk were consistent with previous other studies (Cardello et al., 1999; Fujimaru et al., 2012; Kim et al., 2015; Wee et al., 2018; EFSA Panel on Food Additives and Flavourings (FAF) et al., 2019; Jung et al., 2021). Similar power function results were found by (Li et al., 2015b; a) where sweeteners in water had a higher power function slope than in chocolate milk, and by Drake et al. (2011) with salt addition in water versus soup or cheese sauce. Similar power function results for sucralose, stevia and monk fruit were observed in skim chocolate milk, whey protein, and yogurt for these nonnutritive sweeteners (Reis et al., 2011; Li et al., 2015a; Wagoner et al., 2018). The relative sweetness of allulose in chocolate milk is consistent with (Jiang et al., 2020) that also found allulose as less intense in sweetness than sucrose. When mixed with sucrose in water, allulose exhibited similar sweetening characteristics similar to sucrose using general labelled magnitude scaling (Wee et al., 2018).

Chocolate milks sweetened with monk fruit, stevia and sucralose exhibited bitter and metallic tastes by descriptive analysis and TDS. Previous studies have observed similar results in various food products containing these nonnutritive sweeteners (DuBois and Lee, 1983; Cardello et al., 1999; Reis et al., 2011; Li et al., 2015a; Keefer et al., 2020). The differences in adult consumer liking among the chocolate milks with different sweeteners in experiment 1 is likely due to the presence of bitter and metallic tastes or other temporal differences that have been documented among various sweeteners compared to sucrose at iso sweet concentration (Reis et al., 2011; Wee et al., 2018; Tan et al., 2019). (Li et al., 2015a) reported that chocolate milks

sweetened with stevia or monk fruit were not acceptable by adults consumers and were significantly lower ($p<0.05$) in overall and aftertaste liking compared to the sucrose control chocolate milk, consistent with the results for the chocolate milks sweetened with stevia or monk fruit in the current study.

Allulose sweetened chocolate milk was not different from the sucrose control chocolate milk, most likely due to the similar sweetness profile with no off-flavors or side tastes evident by descriptive analysis, TDS and adult consumer testing (Hossain et al., 2015; Wee et al., 2018). (Mora et al., 2022) also found similar results in adult consumer liking and purchase intent between allulose and sucrose sweetened yogurts. (Tan et al., 2019) reported a low citation of bitterness with allulose compared to sucrose in water, however, their results were generated with higher concentrations of sucrose and allulose than the current study. Overall, allulose has exhibited similar sensory profiles to sucrose through descriptive analysis, temporal sensory profiling and consumer testing.

Results from this study indicates that lactose hydrolysis can reduce the amount of added sweetener to chocolate milk while maintaining sweetness equivalence. (Li et al., 2015b) reported hydrolysis of >7.5% (wt/vol) of added lactose in chocolate milk achieved similar sweetness profile to 150 n*M* sucrose chocolate milk, and no differences in overall liking and sweetness liking ($p>0.05$) were detected among young adult consumers. Even though the >7.5% added lactose achieved sweetness equivalence to the sucrose chocolate milk, the added lactose at that level was too high to fulfill a calorie-reduced requirement, unlike the current study where the existing lactose was hydrolyzed to increase the sweetness of the milk without contributing calories. Previous studies have indicated that lactose free milk via hydrolysis is sweeter than regular milk, and hydrolysis of 70% of existing lactose in milk can increase the sweetness of

dairy products to the same degree as adding 2% sucrose (Zadow, 1986; Mahoney, 1998; Adhikari et al., 2010; McCain et al., 2018). (Whalen et al., 1988) reported consumers could not detect a difference between yogurt sweetened with sugar (4 g of sucrose/100 g of yogurt) and lactose-hydrolyzed yogurt with less added sugar (2-3 g of sucrose/100 g of yogurt). The sensory differences among chocolate milks in experiment 2 compared to experiment 1 were smaller by using lactose hydrolysis to sweeten the chocolate milk base and thus use less added sweetener. Using lactose-hydrolyzed milk with nonnutritive sweeteners can achieve a similar sweetness profile to sucrose chocolate milk with no added sugar or calories.

The use of sweetener blends improved the sensory profiles of the chocolate milks to resemble the flavor profile of sucrose. In the current study, chocolate milks with nonnutritive sweetener blends scored at parity with the sucrose control chocolate milk. (Parker et al., 2018) also found that sweetener blends of monk fruit and stevia closely matched the consumer acceptance scores of sucrose-sweetened vanilla RTM protein beverages for overall liking and flavor liking. This was evident in experiment 3 of the current study that noted only minor differences among sweetener blends in LH chocolate milks and no differences in liking by adults or children. (Li et al., 2015a) reported metallic and bitter taste in chocolate milks sweetened with stevia: sucrose and monk fruit: sucrose blends in chocolate milks when compared to the control 5.14% sucrose chocolate milk. These results may be due to differences in the sources/purity of stevia and monk fruit used in their study or due to higher concentrations of the sweeteners themselves that were used in their study. Using sweetener blends can reduce the cost to produce, as natural nonnutritive sweeteners are higher in cost compared to sucrose. As a blend, stevia can reduce the total cost while replicating the sweetness profile of sucrose without contributing any calories (see Appendix F).

When selecting beverages for their children, (Schiano et al., 2022) identified that parents valued themes including no high sugar or added sugar, non-soda beverage, healthy, natural/not artificial, nutrient content, and something kid's would enjoy. Racette et al. (2023) reported that the ideal school lunch milk for parents was unflavored or chocolate 2% fat, ultrapasteurized milk with all-natural and hormone-free label claims. This study also noted that parents were unfamiliar with the specific attributes or nutritional profile of milk served in schools, but believed milk should be served as part of school breakfast and lunch, and valued flavored milk as a vehicle for delivering important nutrients for their children. A lactose free school lunch milk was also appealing to parents (Racette et al., 2023).Lactose hydrolyzed milks with stevia, LH blend #2, and LH blend #4 fit the themes that parents valued, while also scoring at parity for all liking attributes by children to the sucrose control chocolate milk. (Yoo et al., 2017) noted that children respond positively to sugar reduction claims among dairy products, and (Sipple et al., 2021) demonstrated that school age children also conceptually wanted milk with superior nutritional quality (e.g. sugar reduced) while also retaining an acceptable flavor. Based on results from the current study, zero-added sugar lactose free chocolate milks could be acceptable among school aged children and their parents. These study results provide novel insights regarding the use of allulose in sweetener blends in chocolate milk, and the use of lactose hydrolysis for chocolate milk to obtain reduction in added sugar content while retaining sweetness.

A limitation of this study was that the respondents were generally from Raleigh, NC and may not represent adult and child milk consumers nationally. A limitation to the applications of this study is that allulose is currently an expensive sweetener and has a low relative sweetness compared to stevia or monk fruit and coupled with GRAS limitations on its usage is not a viable option for use as the sole sweetener in school lunch chocolate milk. Another limitation to

consider in this study is the use of the specific types of monk fruit and stevia used. Monk fruit and stevia consist of different glycosides in varying purity and proportions and different sources may have distinct sensory profiles.

Conclusion

This study evaluated the application of nonnutritive sweetener blends and lactose hydrolysis to replicate the sensory profile of 3.69% (w/v) sucrose-sweetened school lunch chocolate milk to minimize calorie content and added sugar. The application of lactose hydrolysis resulted in an appealing lactose free claim but also reduced the amount of each sucrose alternative sweetener/sweetener blend needed to reach isosweetness, resulting in similar temporal sensory profiles and no differences ($p>0.05$) in overall liking to the sucrose control chocolate milk among adult and child consumers. A combination of monk fruit, stevia and/or allulose with lactose-hydrolysis can produce chocolate milk with similar sensory profiles to sucrose. The findings from this can be useful for milk manufacturers in accomplishing the objective of a school lunch milk that is appealing to parents and children. Future work on lactose-hydrolyzed milk beverages using nonnutritive sweetener blends to reduce added sugar content while retaining sweetness profile should be further researched.

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Tables and Figures

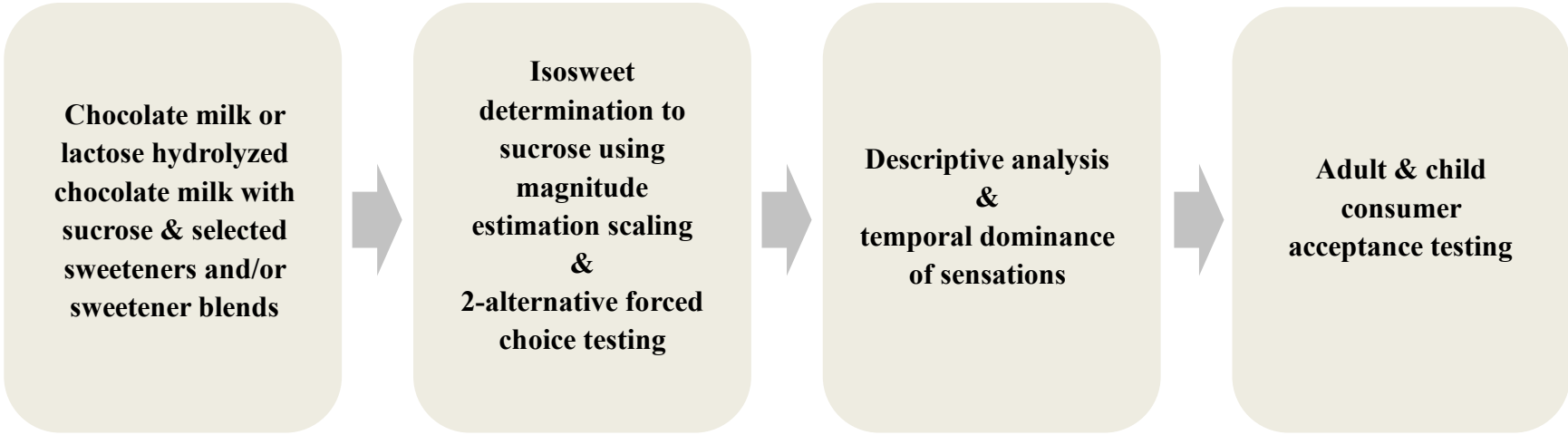


Figure 1. *Experimental overview*

Table 1. Formulations of 1% milkfat chocolate milks in experiment 1 (per 1000 g batch)

Sample ID	Sucrose (g)	Allulose (g)	Mixed sugar syrup (g)	Monk fruit extract (mg)	Stevia leaf extract (mg)	Sucralose (mg)	Chocolate milk base (g)
Sucrose control	36.9	-	-	-	-	-	963.1
Allulose	-	105.7	-	-	-	-	894.3
Mixed sugar syrup	-	-	65.3	-	-	-	934.7
Monk fruit	-	-	-	258.4	-	-	999.7
Stevia	-	-	-	-	205.3	-	999.7
Sucralose	-	-	-	-	-	64.9	999.9

Chocolate milk with sweeteners were formulated to sweetness equivalency 3.69% w/w sucrose in chocolate milk.

Table 2. Formulations of 1% milkfat lactose hydrolyzed (LH) chocolate milks in experiment 2 (per 1000 g batch)

Sample ID	Sucrose (g)	Allulose (g)	Mixed sugar syrup (g)	Monk fruit extract (mg)	Stevia leaf extract (mg)	Chocolate milk base (g)
Sucrose Control	36.9	-	-	-	-	963.1
LH Sucrose	25.3	-	-	-	-	974.7
LH Allulose	-	71.0	-	-	-	929.0
LH Mixed sugar syrup	-	-	40.9	-	-	959.1
LH Monk fruit	-	-	-	192.2	-	999.8
LH Stevia	-	-	-	-	143.1	999.9

Chocolate milk with sweeteners were formulated to sweet taste equivalency 3.69% w/w sucrose in chocolate milk.
LH: Lactose hydrolyzed.

Table 3. Formulations of 1% milkfat lactose hydrolyzed (LH) chocolate milks in experiment 3 (per 1000 g batch)

Sample ID	Sucrose (g/L)	Allulose (g)	Monk fruit extract (mg)	Stevia leaf extract (mg)	Chocolate milk base (g)
Sucrose control	36.9	-	-	-	963.1
LH Sucrose	25.3	-	-	-	974.7
LH Blend #1	-	-	96.1	71.6	999.9
LH Blend #2	-	14.0	70.0	52.0	986.0
LH Blend #3	-	27.2	78.0	-	972.8
LH Blend #4	-	-	48.1	107.3	999.9

[1]Chocolate milks were formulated to sweet taste equivalency of 3.69% w/w sucrose in chocolate milk.
LH: Lactose hydrolyzed.
LH Blend #1 = 50% stevia, 50% monk fruit; LH Blend #2 = 40% stevia, 40% monk fruit, 20% allulose; LH Blend #3 = 60% monk fruit, 40% allulose; LH Blend #4 = 75% stevia, 25% monk fruit.

Table 4. Formulations of 1% milkfat chocolate milks with or without lactose hydrolysis in children's acceptance test in experiment 3 (per 1000 g batch)

Sample ID	Sucrose (g)	Allulose (g)	Monk fruit extract (mg)	Stevia leaf extract (mg)	Chocolate milk base(g)	Grams of added sugar/240 mL serving	Calories from sweetener/ 240mL serving
Sucrose control	36.9	-	-	-	963.1	8.9	35.4
LH Sucrose	25.3	-	-	-	974.7	6.1	24.5
LH Stevia	-	-	-	143.1	999.9	0.0	0.0
LH Blend #2	-	14.0	70.0	52.0	986.0	0.0	1.4
LH Blend #4	-	-	48.1	107.3	999.9	0.0	0.0

[1]Chocolate milks were formulated to sweet taste equivalency of 3.69% sucrose in chocolate milk
LH: Lactose hydrolyzed.
LH Blend #2 = 40% stevia, 40% monk fruit, 20% allulose; LH Blend #4 = 75% stevia, 25% monk fruit.

Table 5. Iso-sweet concentration of sweeteners in chocolate milk to 3.69% sucrose in 1% milkfat chocolate milks in experiment 1

	Iso-sweet concentration	Relative sweetness[1]	Calories (kcal)/ gram of sweetener	Added sweetener (g)[3]	Calories from added sugar/240 mL serving (kcal)[4]
Sucrose Control	3.69%	1.0	4.0	8.9	35.4
Allulose	10.57%	0.4	0.4	25.4	10.1
Mixed Sugar Syrup	6.53%	0.6	2.0	15.7	31.3
Monk Fruit	0.03%	143.0	0.0	0.1	0.0
Stevia	0.02%	180.0	0.0	0.1	0.0
Sucralose	0.01%	568.0	0.0	0.0	0.0

[1]Relative sweetness to sucrose represents the measure of how sweet the nutritive/nonnutritive sweetener is in relation to sucrose-sweetened chocolate milk.
[2]Calories/gram of sweetener is the amount of calories (kcal) per gram (g) of sweetener used in chocolate milk
[3]Added sweetener indicates the amount (g) of sweetener per 240 mL serving at the iso-sweet concentration of 3.69% w/w sucrose in chocolate milk.
[4]Calories from added sugar indicates the amount of calories from added sweetener per 240mL serving of chocolate milk at the iso-sweet concentration of 3.69% sucrose in chocolate milk.

Table 6. Power function slopes of sweeteners in water, chocolate milk and lactose hydrolyzed chocolate milk

	Water	Chocolate Milk	LH Chocolate Milk
Allulose	1.484Aa	1.066Ab	0.931ABb
Mixed Sugar Syrup	1.097BCa	0.899Bb	0.994Ab
Monk Fruit	0.935BCa	0.802CDb	0.811Cb
Stevia	1.031BCa	0.855BCb	0.870BCb
Sucralose	1.154Ba	0.775CDb	-
Sucrose	0.911Ca	0.730Db	0.857Cab

Slopes with different lowercase lettering within rows represents significant differences ($p<0.05$) among liquid solutions with the same sweetener. Slopes with different uppercase lettering within columns represents significant differences ($p<0.05$) among sweeteners in the same liquid solution

Table 7. Descriptive analysis means for 1% milkfat chocolate milks sweetened with nutritive and nonnutritive sweeteners (experiment 1)

Sample ID	Overall aroma	Cocoa flavor	Cooked/ Milky	Cardboard	Sweet	Salty	Bitter	Metallic	Astringent	Viscosity
Sucrose control	1.9bc	1.9a	3.0a	0.6a	5.8b	1.8a	ND	ND	2.1a	2.1a
Allulose	1.8c	2.1a	3.2a	0.6a	5.9ab	1.7a	ND	ND	1.9b	1.9a
Mixed sugar syrup	2.0ab	1.9a	2.9a	0.5a	6.0a	1.7a	ND	ND	2.0ab	2.0a
Monk fruit	2.2a	1.9a	3.0a	0.6a	5.8b	1.9a	0.8a	1.0a	1.9b	2.1a
Stevia	1.8c	2.1a	3.1a	0.5a	5.8b	1.9a	1.0a	1.0a	2.0ab	1.9a
Sucralose	1.9bc	2.0a	2.9a	0.5a	5.9ab	1.9a	ND	1.0a	2.0ab	2.0a

Values represent the mean scores from duplicate panelist evaluations (n=8). Attribute intensities were scored on a 0 to 15-point universal intensity scale (Meilgaard et al., 2007). ND – not detected. Means within a column followed by a different letter represent differences ($P < 0.05$).

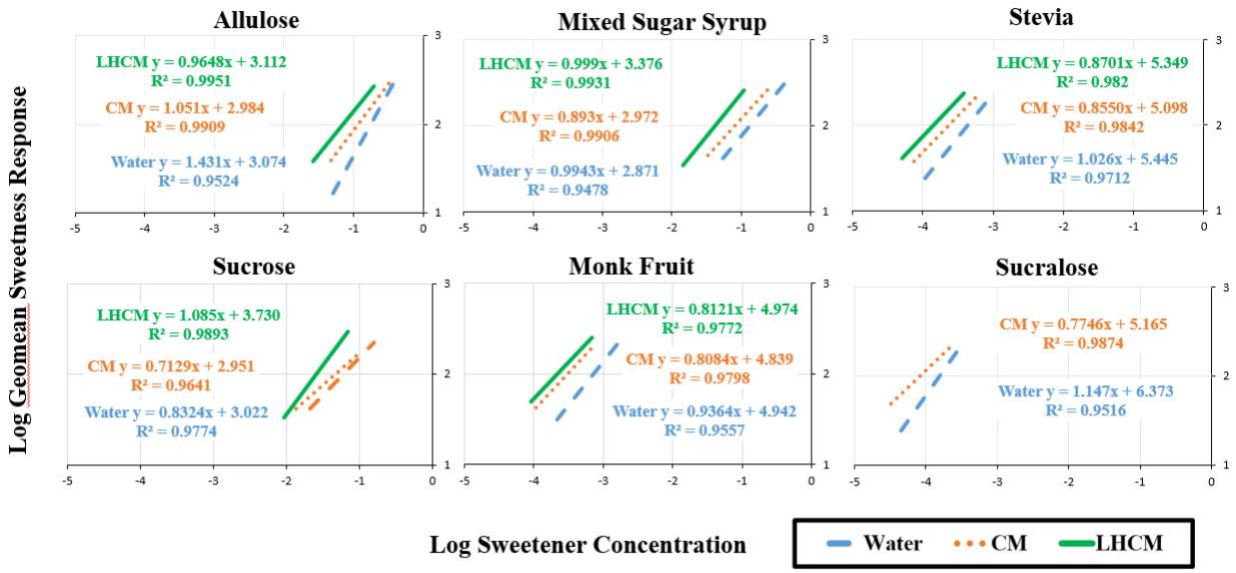


Figure 2. *Sweetness power function slopes of nutritive and nonnutritive sweeteners from experiments I (water and chocolate milk) and II (lactose hydrolyzed chocolate milk) in water, chocolate milk and lactose hydrolyzed chocolate milk matrices*. CM: chocolate milk. LHCM: Lactose hydrolyzed chocolate milk. Power function are represented as the best-fit line of three independent replications across seven sweetener concentrations for each nutritive and nonnutritive sweetener. The y=2 axis indicates the scaled sweetness intensity of the current school lunch chocolate milk formulation used as the reference 100 (Wagoner et al., 2018).

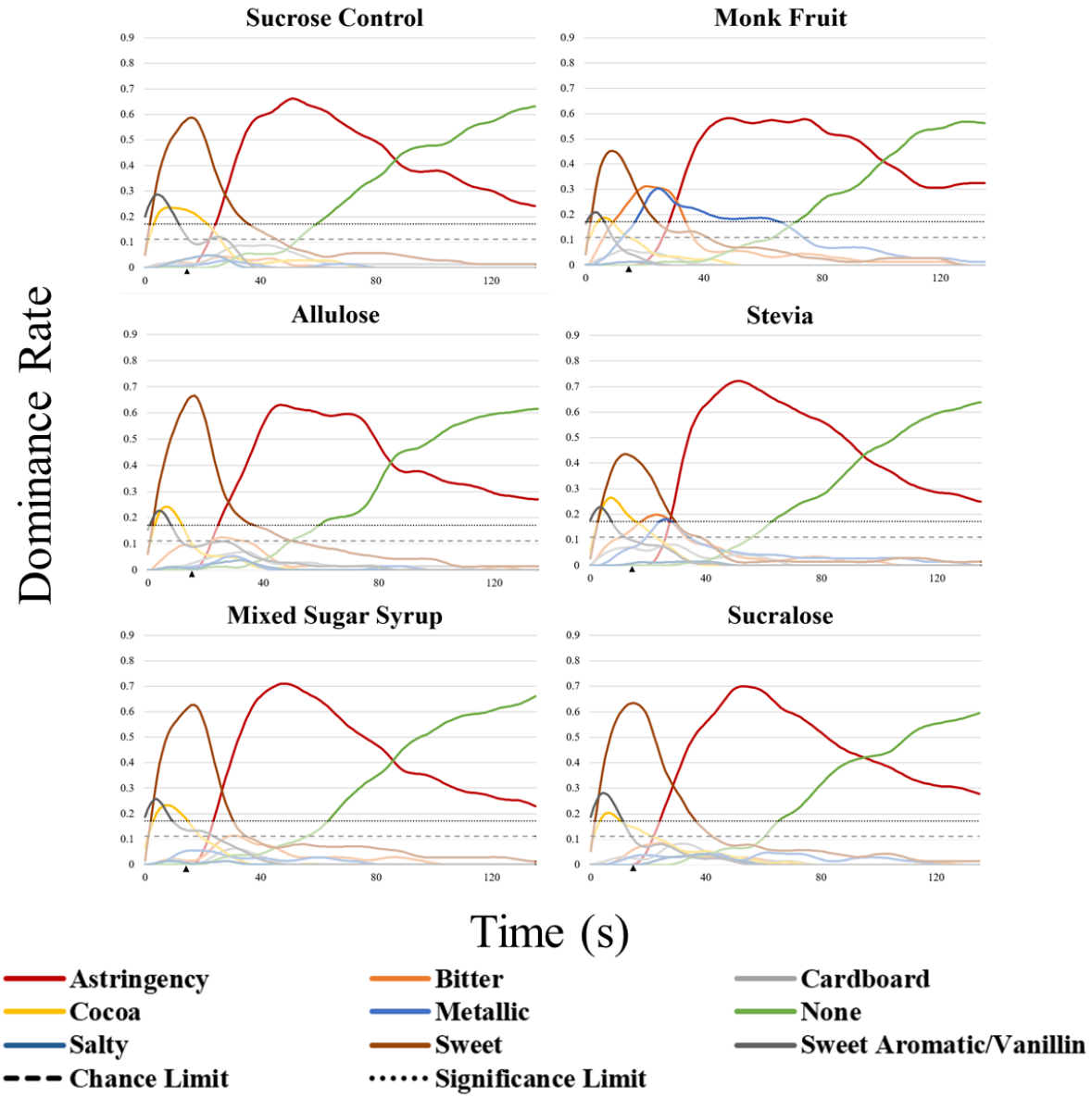


Figure 3. *Experiment 1 temporal dominance of sensations frequency of attributes for sucrose control, allulose, mixed sugar syrup, monk fruit, stevia and sucralose in chocolate milk*. Dotted line indicates significance level ($\alpha = 0.05$) and dashed line indicates chance limit. The triangle on the x-axis is located at x=15 seconds to indicate the time when samples were expectorated. Data below the significance line is faded to facilitate comparisons among samples. The chance limit is the dominance rate that an attribute can obtain by chance and is defined by P0=1/K, K being the number of attributes. The significance limit indicates when an attribute is significantly dominant at a specific time.

Table 8. Adult consumer acceptance scores of 1% milkfat chocolate milks (experiment 1) (n=122)

		Sucrose Control	Allulose	Mixed Sugar Syrup	Monk Fruit	Stevia	Sucralose
Appearance Liking		6.6a	6.8a	6.6a	6.6a	6.6a	6.6a
Color Liking		6.5a	6.6a	6.5a	6.5a	6.5a	6.5a
Aroma Liking		6.0a	5.8a	5.8a	5.7a	5.8a	5.8a
Overall Liking		6.3a	5.9ab	6.2ab	5.3c	5.8b	5.9ab
Overall Flavor Liking		6.1a	5.7ab	6.1a	5.1c	5.5bc	5.7ab
Overall Flavor JAR		6.1a	5.7ab	6.1a	5.1c	5.5bc	5.7ab
Sweetness Liking		6.3a	5.7bc	6.1ab	4.9d	5.5c	5.5c
Chocolate Flavor Liking		5.9a	5.8ab	5.9ab	5.4b	5.5ab	5.6ab
Mouthfeel/Thickness Liking		6.4a	6.3a	6.4a	6.1a	6.1a	6.3a
Aftertaste Liking		5.7a	4.4cd	5.3ab	3.8d	3.8d	4.6bc
Color JAR	Too Light	25.4%a	**32.0%a**	**27.9%a**	**37.7%a**	**36.1%a**	**33.6%a**
	Just About Right	73.0%a	66.4%a	68.9%a	61.5%a	63.9%a	63.9%a
	Too Dark	1.6%a	1.6%a	3.3%a	0.8%a	0.0%a	2.5%a
Aroma JAR	Too Weak	**48.4%a**	53.3%a	**50.8%a**	54.9%a	53.3%a	51.6%a
	Just About Right	47.5%a	38.5%a	45.9%a	39.3%a	42.6%a	45.1%a
	Too Strong	4.1%a	8.2%a	3.3%a	5.7%a	4.1%a	3.3%a
Overall Flavor	Too Weak	**38.5%a**	**42.6%a**	**37.7%a**	**38.5%a**	**36.9%a**	**37.7%a**
	Just About Right	56.6%a	45.9%a	53.3%a	36.1%a	45.1%a	50.0%a
	Too Strong	4.9%b	11.5%ab	9.0%b	**25.4%a**	18.0%ab	12.3%ab
Sweetness JAR	Not Sweet Enough	15.6%a	**24.6%a**	**22.1%a**	**23.0%a**	16.4%a	16.4%a
	Just About Right	70.5%a	59.8%ab	64.8%ab	38.5%c	50%bc	45.9%bc
	Too Sweet	13.9%b	15.6%b	13.1%b	**38.5%a**	**33.6%a**	**37.7%a**

Table 9. Adult consumer acceptance scores of 1% milkfat chocolate milks (experiment 1) (n=122) (continued)

Chocolate Flavor JAR	Too Weak	**49.2%a**	**50.8%a**	**50.8%a**	**54.1%a**	**50.8%a**	**57.4%a**
	Just About Right	47.5%a	42.6%a	45.1%a	39.3%a	41.8%a	37.7%a
	Too Strong	3.3%a	6.6%a	4.1%a	6.6%a	7.4%a	4.9%a
Mouthfeel/ Thickness JAR	Too Thin	28.7%a	**31.1%a**	**25.4%a**	25.4%a	**29.5%a**	**27.9%a**
	Just About Right	67.2%a	66.4%a	69.7%a	67.2%a	63.9%a	66.4%a
	Too Thick	4.1%a	2.5%a	4.9%a	7.4%a	6.6%a	5.7%a
Aftertaste Y/N?	Yes	27.0%c (n=33)	33.6%bc (n=41)	31.1%bc (n=38)	54.9%a (n=67	50.8%ab (n=62)	42.6%abc (n=52)
	No	73.0%a (n=89)	66.4%ab (n=81)	68.9%ab (n=84	45.1%c (n=55)	49.2%bc (n=60)	57.4%abc (n=70)
Aftertaste JAR	Too Weak	9.1%a	0.0%a	10.5%a	9.0%a	1.6%a	5.8%a
	Just About Right	60.6%a	46.3%ab	57.9%a	23.9%bc	17.7%c	46.2%ab
	Too Strong	**30.3%c**	**53.7%abc**	31.6%c	**67.2%ab**	**80.6%a**	**48.1%bc**
Aftertaste Pleasantness		3.3a	2.6abc	2.9a	2.2c	2.2bc	2.8ab
Expectations		2.9a	2.6ab	2.8a	2.3c	2.5bc	2.7ab
Purchase Intent		3.0a	2.7ab	3.0a	2.3b	2.5b	2.7ab

Means within a row followed by a different letter represent significant differences ($p<0.05$)
Liking questions were scored on a 9-point hedonic scale where dislike extremely=1 and like extremely=9
JAR questions were scored on a 5-point scale where 1 or 2 = too little, 3 = just about right, and 4 or 5 = too much
Aftertaste Liking, Aftertaste Pleasantness, and Aftertaste JAR were asked to consumer who could detect an aftertaste flavor (Yes/No)
Aftertaste Pleasantness was scored on a 5-point scale where 1-2= unpleasant, 3= neither pleasant nor unpleasant, and 4-5= pleasant. Statistical lettering was determined using Kruskal-Wallis testing
Expectations was scored on a 5-point scale where 1-2=did not meet expectations, 3= meets expectations, and 4-5=exceeds expectations. Statistical lettering was determined using Kruskal-Wallis testing
Purchase Intent was scored on a 5-point scale where 1-2=would not purchase, 3= maybe/maybe not purchase, and 4-5=would purchase. Statistical lettering was determined using Kruskal-Wallis testing
Bolded percentages for JAR question attributes received significant penalty ($p<0.05$)

Table 10. Iso-sweet concentration of sweeteners in 1% milkfat lactose hydrolyzed chocolate milk to 3.69% sucrose in 1% milkfat chocolate milk in experiment 2

Sample ID	Iso-sweet concentration[1]	Relative sweetness[2]	Added sweetener (g)[4]	Calories from added sugar (kcal)[5]
Sucrose Control	3.69%	1.0	8.9	35.4
LH Sucrose	2.53%	1.5	6.1	10.1
LH Allulose	7.10%	0.5	17.0	6.8
LH Mixed sugar syrup	4.09%	0.9	9.8	19.6
LH Monk fruit	0.02%	192.2	0.05	0.0
LH Stevia	0.01%	258.0	0.03	0.0

LH: lactose hydrolyzed.

[1]Iso-sweet concentration indicates sweetness equivalence concentration of nutritive and nonnutritive sweeteners in chocolate milk to 3.69% w/w sucrose in 1% milkfat chocolate milk solution

[2]Relative sweetness is a ratio comparing the iso-sweet concentration of sucrose with a sweetener.

[3]Calories/gram of sweetener is the amount of calories (kcal) per gram (g) of sweetener used in chocolate milk

[4]Added sweetener indicates the amount (g) of sweetener per 240 mL chocolate milk serving at the iso-sweet concentration.

[5]Calories from added sugar indicates additional calories (kcal) due to added sweetener per 240 mL serving of chocolate milk at iso-sweet concentration

Table 11. Descriptive analysis means for 1% milkfat lactose hydrolyzed chocolate milks sweetened with nutritive and nonnutritive sweeteners (experiment 2)

Sample ID	Overall aroma	Cocoa flavor	Cooked/ Milky	Cardboard	Sweet	Salty	Bitter	Metallic	Astringent	Viscosity
LH Sucrose	2.0c	2.1c	3.2a	0.5b	5.9a	1.8b	ND	ND	1.8c	2.2a
LH Allulose	2.3a	2.0b	3.3a	0.6a	5.8a	2.0b	ND	ND	2.0b	2.1b
LH Mixed sugar syrup	2.3a	2.0b	3.0b	0.5b	5.9a	2.2a	ND	ND	1.8c	2.2a
LH Monk fruit	2.0c	2.3a	3.3a	0.5b	5.9a	1.8d	ND	0.5a	2.2a	2.1b
LH Stevia	2.2c	2.2a	3.3a	0.7a	5.8a	1.9c	0.5a	0.5a	2.2a	2.1b

Values represent the mean scores from duplicate panelist evaluations (n=8). Attribute intensities were scored on a 0 to 15-point universal intensity scale (Meilgaard et al., 2007). ND – not detected. Means within a column followed by a different letter represent differences ($P < 0.05$).
LH: Lactose hydrolyzed

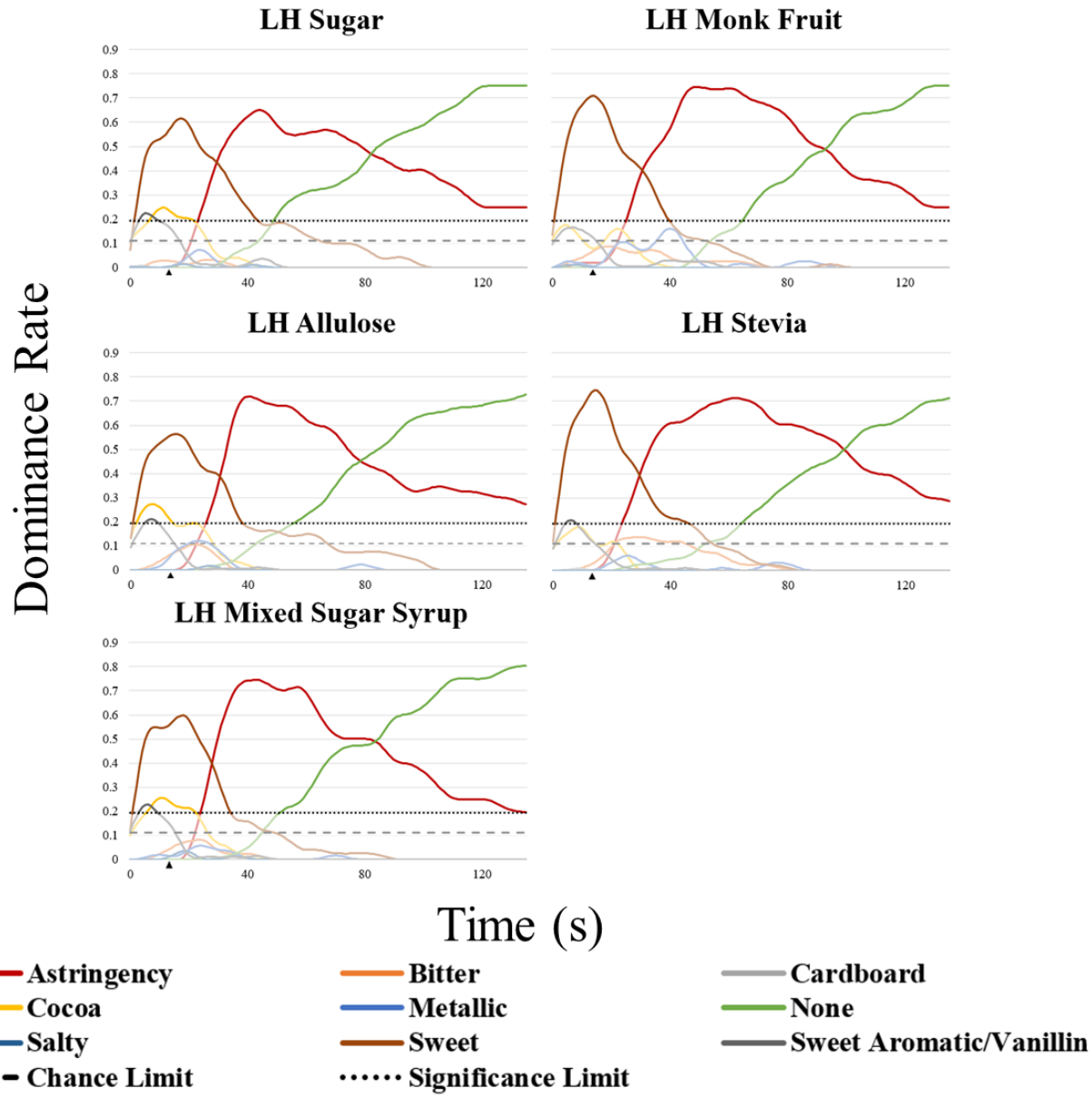


Figure 4. *Experiment 2 temporal dominance of sensations frequencies of attributes of 1% milkfat lactose hydrolyzed (LH) chocolate milks with sucrose, allulose, mixed sugar syrup, monk fruit and stevia*. Dotted line indicates significance level ($\alpha = 0.05$) and dashed line indicates chance limit. The triangle on the x-axis is located at x=15 seconds to indicate the time when samples were expectorated. Data below the significance line is faded to facilitate comparisons among samples. The chance limit is the dominance rate that an attribute can obtain by chance and is defined by P0=1/K, K being the number of attributes. The significance limit indicates when an attribute is significantly dominant at a specific time.

Table 12. Adult consumer acceptance scores of 1% milkfat lactose hydrolyzed chocolate milk (experiment 2) (n=160)

		Sucrose Control	LH Sugar	LH Allulose	LH MSS	LH Monk Fruit	LH Stevia
Appearance Liking		6.9a	6.7a	6.9a	6.7a	6.8a	6.9a
Color Liking		6.6a	6.5a	6.7a	6.6a	6.6a	6.8a
Aroma Liking		6.3a	6.3a	6.2a	6.3a	6.6a	6.2a
Overall Liking		6.6a	6.5a	6.5a	6.3a	6.4a	6.5a
Overall Flavor Liking		6.5a	6.3a	6.2a	6.2a	6.1a	6.3a
Sweetness Liking		6.6a	6.5a	6.3a	6.4a	6.1a	6.1a
Chocolate Flavor Liking		6.3ab	6.1abc	6.0bc	5.8c	6.4a	6.3ab
Mouthfeel/Thickness Liking		6.7a	6.7a	6.6a	6.7a	6.5a	6.6a
Aftertaste Liking		5.8a	5.6ab	4.9bc	5.3ab	4.6c	4.9bc
Color JAR	Too Light	**45.0%a**	**43.8%a**	**36.3%a**	**43.8%a**	43.1%a	39.4%a
	Just About Right	52.5%a	55.6%a	63.1%a	55%a	55%a	60.6%a
	Too Dark	2.5%a	0.6%a	0.6%a	1.3%a	1.9%a	0.0%a
Aroma JAR	Too Weak	**50.6%a**	**48.8%a**	51.9%a	**47.5%a**	40.6%a	45.6%a
	Just About Right	46.3%a	49.4%a	45.6%a	49.4%a	55.6%a	50.0%a
	Too Strong	3.1%a	1.9%a	2.5%a	3.1%a	3.8%a	4.4%a
Overall Flavor JAR	Too Weak	**39.4%abc**	**44.4%ab**	**41.3%abc**	**53.1%a**	**28.8%bc**	**24.4%c**
	Just About Right	55.0%a	50.0%a	52.5%a	43.1%a	57.5%a	58.1%a
	Too Strong	5.6%b	5.6%b	6.3%ab	3.8%b	13.8%ab	17.5%a
Sweetness JAR	Not Sweet Enough	**20.0%ab**	**21.3%ab**	**26.9%ab**	**30.0%a**	13.1%b	13.8%b
	Just About Right	63.8%a	66.3%a	62.5%a	62.5%a	53.8%a	55.6%a
	Too Sweet	16.3%bc	12.5%c	10.6%c	7.5%c	**33.1%a**	**30.6%ab**
Chocolate Flavor JAR	Too Weak	**51.3%ab**	**55.0%ab**	**53.8%ab**	**59.4%a**	**36.9%b**	**37.5%b**
	Just About Right	45.6%a	41.9%a	43.1%a	37.5%a	55.0%a	53.8%a
	Too Strong	3.1%a	3.1%a	3.1%a	3.1%a	8.1%a	8.8%a
Mouthfeel/ Thickness JAR	Too Thin	**21.3%a**	**21.3%a**	**20.0%a**	19.4%a	**21.9%a**	15.0%a
	Just About Right	74.4%a	73.8%a	72.5%a	72.5%a	68.8%a	75.0%a
	Too Thick	4.4%a	5.0%a	7.5%a	8.1%a	9.4%a	10.0%a

Table 13. Adult consumer acceptance scores of 1% milkfat lactose hydrolyzed chocolate milk (experiment 2) (n=160) (continued)

Aftertaste Y/N?	Yes	34.40% (n=55)	31.90% (n=51)	40.60% (n=65)	35.60% (n=57)	51.30% (n=82)	46.90% (n=75)
	No	65.60% (n=105)	68.10% (n=109)	59.40% (n=95)	64.40% (n=103)	48.80% (n=78)	53.10% (n=85)
Aftertaste JAR	Too Weak	9.1%a	5.9%a	10.8%a	8.8%a	4.9%a	5.3%a
	Just About Right	58.2%ab	62.7%a	49.2%ab	52.6%ab	34.1%b	52.0%ab
	Too Strong	32.7%b	31.4%b	**40.0%ab**	**38.6%ab**	**61.0%a**	**42.7%ab**
Aftertaste Pleasantness		3.2a	3.1ab	3.1ab	3.0abc	2.6d	2.6cd
Expectations		3.0a	3.0.a	3.0.a	2.8b	2.8ab	2.8ab
Purchase Intent		3.1a	3.0a	3.0a	2.9ab	2.7b	2.9ab

Means within a row followed by a different letter represent significant differences ($p<0.05$)
Liking questions were scored on a 9-point hedonic scale where dislike extremely=1 and like extremely=9
JAR questions were scored on a 5-point scale where 1 or 2 = too little, 3 = just about right, and 4 or 5 = too much
Aftertaste Liking, Aftertaste Pleasantness, and Aftertaste JAR were asked to consumer who could detect an aftertaste flavor (Yes/No)
Aftertaste Pleasantness was scored on a 5-point scale where 1-2= unpleasant, 3= neither pleasant nor unpleasant, and 4-5= pleasant. Statistical lettering was determined using Kruskal-Wallis testing
Expectations was scored on a 5-point scale where 1-2=did not meet expectations, 3= meets expectations, and 4-5=exceeds expectations. Statistical lettering was determined using Kruskal-Wallis testing
Purchase Intent was scored on a 5-point scale where 1-2=would not purchase, 3= maybe/maybe not purchase, and 4-5=would purchase. Statistical lettering was determined using Kruskal-Wallis testing
Bolded percentages for JAR question attributes received significant penalty ($p<0.05$)

Table 14. Descriptive analysis means for nonnutritive sweetener blends in 1% milkfat lactose hydrolyzed chocolate milks (experiment 3).

Sample	Overall aroma	Cocoa flavor	Cooked/ Milky	Cardboard	Sweet	Salty	Bitter	Metallic	Astringent	Viscosity
LH Blend #1	2.5b	2.2a	3.1c	0.6b	6.0b	1.4b	ND	ND	1.6a	1.8c
LH Blend #2	2.4c	2.2a	3.3a	0.5b	6.1a	1.5a	ND	ND	1.5a	2.1a
LH Blend #3	2.6a	2.2a	3.2b	0.5b	6.0b	1.5a	ND	ND	1.6a	2.0b
LH Blend #4	2.5b	2.1ab	3.2b	0.8a	6.0b	1.4b	ND	ND	1.4b	2.0b

Values represent the mean scores from duplicate panelist evaluations (n=9). Attribute intensities were scored on a 0 to 15-point universal intensity scale (Meilgaard et al., 2007). ND – not detected. LH: Lactose hydrolyzed. chocolate milk: School lunch chocolate milk.
Means within a column followed by a different letter represent differences ($P < 0.05$).
LH Blend #1 = 50% stevia, 50% monk fruit; LH Blend #2 = 40% stevia, 40% monk fruit, 20% allulose; LH Blend #3 = 60% monk fruit, 40% allulose; LH Blend #4 = 75% stevia, 25% monk fruit.
LH: Lactose hydrolyze

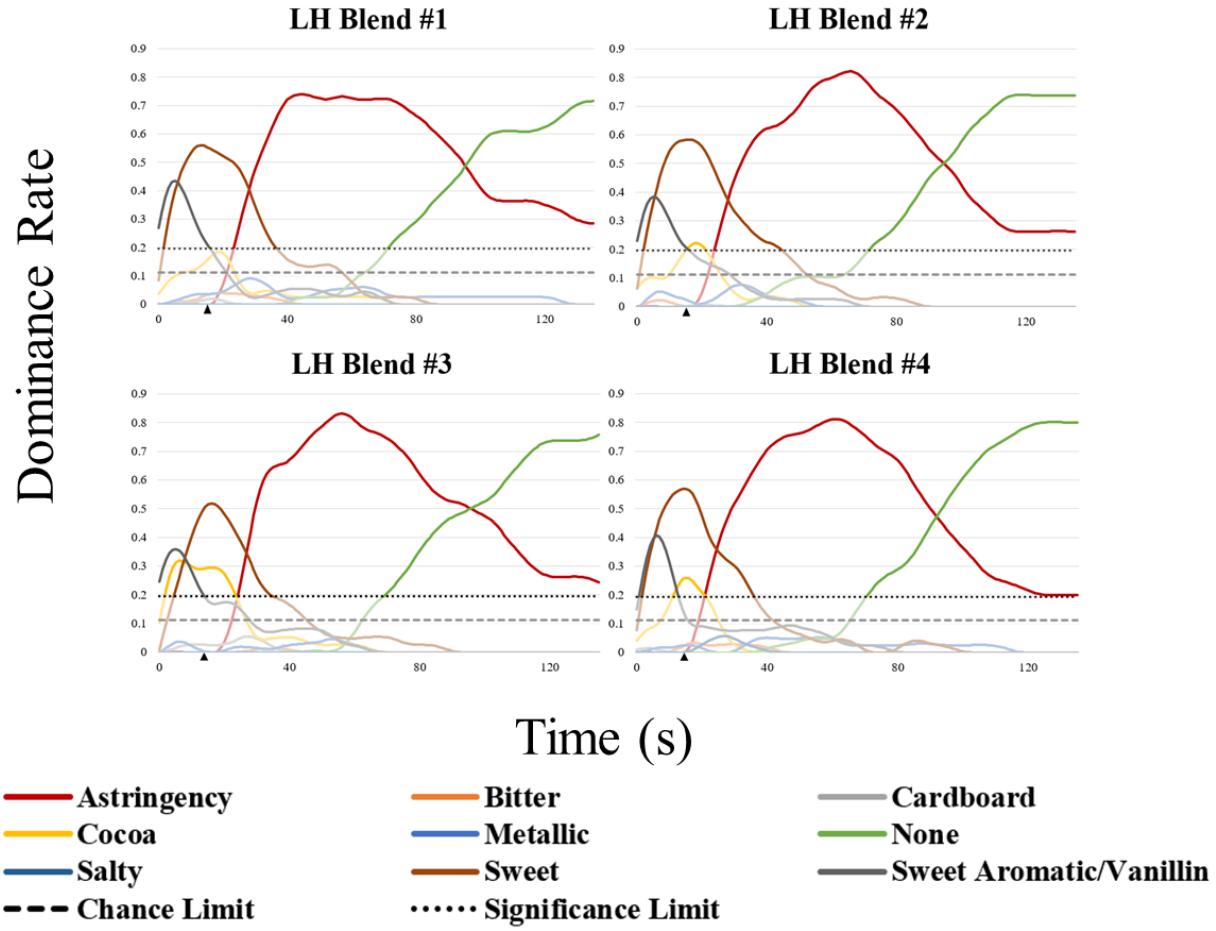


Figure 5. *Experiment 3 temporal dominance of sensations frequencies of attributes of 1% milkfat lactose hydrolyzed (LH) chocolate milks with sweetener blends (Table 3) for experiment 3*. Dotted line indicates significance level (α = 0.05) and dashed line indicates chance limit. The triangle on the x-axis is located at x=15 seconds to indicate the time when samples were expectorated. Data below the significance line is faded to facilitate comparisons among samples. The chance limit is the dominance rate that an attribute can obtain by chance and is defined by P0=1/K, K being the number of attributes. The significance limit indicates when an attribute is significantly dominant at a specific time. LH Blend #1 = 50% stevia, 50% monk fruit; LH Blend #2 = 40% stevia, 40% monk fruit, 20% allulose; LH Blend #3 = 60% monk fruit, 40% allulose; LH Blend #4 = 75% stevia, 25% monk fruit.

Table 15. Adult consumer acceptance scores of 1% milkfat lactose hydrolyzed chocolate milk with sweetener blends (experiment 3) (n=131)

		Sucrose Control	LH Sugar	LH Blend #1	LH Blend #2	LH Blend #3	LH Blend #4
Appearance Liking		6.7a	6.7a	6.7a	6.7a	6.7a	6.7a
Color Liking		6.5a	6.5a	6.5a	6.6a	6.5a	6.6a
Aroma Liking		6.5a	6.5a	6.5a	6.5a	6.5a	6.5a
Overall Liking		6.9a	6.6a	6.6a	6.8a	6.5a	6.8a
Overall Flavor Liking		6.7a	6.5a	6.5a	6.5a	6.4a	6.6a
Sweetness Liking		6.8a	6.5ab	6.3b	6.5ab	6.3b	6.4ab
Chocolate Flavor Liking		6.5a	6.1a	6.5a	6.5a	6.2a	6.5a
Mouthfeel/Thickness Liking		6.8ab	6.5ab	6.9a	6.8ab	6.5a	6.7ab
Aftertaste Liking		5.8a	5.4ab	4.8b	5.4ab	5.3b	5.2ab
Color JAR	Too Light	41.2%a	**42.0%a**	47.3%a	**40.5%a**	**42.7%a**	43.5%a
	Just About Right	56.5%a	56.5%a	51.9%a	58.0%a	53.4%a	51.9%a
	Too Dark	2.3%a	1.5%a	0.8%a	1.5%a	3.8%a	4.6%a
Aroma JAR	Too Weak	42.7%a	**38.2%a**	**38.9%a**	**44.3%a**	**39.7%a**	**42.0%a**
	Just About Right	55.7%a	58.8%a	54.2%a	53.4%a	57.3%a	53.4%a
	Too Strong	1.5%a	3.1%a	6.9%a	2.3%a	3.1%a	4.6%a
Overall Flavor JAR	Too Weak	**30.5%ab**	**38.2%ab**	21.4%b	**31.3%ab**	**40.5%a**	**29.0%ab**
	Just About Right	64.1%a	55.0%a	60.3%a	56.5%a	54.2%a	55.7%a
	Too Strong	5.3%a	6.9%a	18.3%a	12.2%a	5.3%a	15.3%a
Sweetness JAR	Not Sweet Enough	12.2%b	**22.9%ab**	9.9%b	19.1%ab	**32.1%a**	13.0%b
	Just About Right	70.2%a	66.4%a	55.0%a	57.3%a	54.2%a	61.8%a
	Too Sweet	17.6%ab	10.7%b	**35.1%a**	**23.7%ab**	13.7%a	**25.2%ab**
Chocolate Flavor JAR	Too Weak	**45.8%a**	**47.3%a**	**35.9%a**	33.6%a	**50.4%a**	**42.7%a**
	Just About Right	50.4%a	48.1%a	54.2%a	55.7%a	45.0%a	51.1%a
	Too Strong	3.8%a	4.6%a	9.9%a	10.7%a	4.6%a	6.1%a
Mouthfeel/Thickness JAR	Too Thin	17.6%a	**25.2%a**	15.3%a	**20.6%a**	**23.7%a**	12.2%a
	Just About Right	76.3%a	63.4%a	75.6%a	70.2%a	64.1%a	72.5%a
	Too Thick	6.1%a	11.5%a	9.2%a	9.2%a	12.2%a	15.3%a

Table 16. Adult consumer acceptance scores of 1% milkfat lactose hydrolyzed chocolate milk with sweetener blends (experiment 3) (n=131) (continued)

Aftertaste Y/N	Yes	35.1% (n=46)	34.4% (n=45)	45.0% (n=59)	38.9% (n=51)	32.1% (n=42)	42.0% (n=55)
	No	64.9% (n=85)	65.6% (n=86)	55.0% (n=72)	61.1% (n=80)	67.9% (n=89)	58.0% (n=76)
Aftertaste JAR	Too Weak	6.5%a	4.4%a	6.8%a	9.8%a	11.9%a	3.6%a
	Just About Right	63.0%a	60.0%a	37.3%a	49.0%a	50.0%a	52.7%a
	Too Strong	30.4%a	35.6%a	**55.9%a**	**41.2%a**	**38.1%a**	**43.6%a**
Aftertaste Pleasantness		3.4a	2.8bc	2.6c	3.1ab	3.0ab	2.9bc
Expectations		3.1a	3.0a	3.0a	3.1a	2.9a	3.0a
Purchase Intent		3.2a	2.9a	3.0a	3.1a	3.0a	3.0a
Purchase Intent Lactose-Free		3.1a	2.9a	2.9a	3.1a	2.9a	3.0a
Purchase Intent Zero Added Sugar		3.2a	3.0a	3.1a	3.2a	3.0a	3.2a
Purchase Intent Lactose Free & Zero Added Sugar		3.1a	3.0a	3.0a	3.2a	2.9a	3.1a

Means within a row followed by a different letter represent significant differences ($p<0.05$)
Liking questions were scored on a 9-point hedonic scale where dislike extremely=1 and like extremely=9
JAR questions were scored on a 5-point scale where 1 or 2 = too little, 3 = just about right, and 4 or 5 = too much
Aftertaste Liking, Aftertaste Pleasantness, and Aftertaste JAR were asked to consumer who could detect an aftertaste flavor (Yes/No)
Aftertaste Pleasantness was scored on a 5-point scale where 1-2= unpleasant, 3= neither pleasant nor unpleasant, and 4-5= pleasant. Statistical lettering was determined using Kruskal-Wallis testing
Expectations was scored on a 5-point scale where 1-2=did not meet expectations, 3= meets expectations, and 4-5=exceeds expectations. Statistical lettering was determined using Kruskal-Wallis testing
Purchase Intent questions was scored on a 5-point scale where 1-2=would not purchase, 3= maybe/maybe not purchase, and 4-5=would purchase. Statistical lettering was determined using Kruskal-Wallis testing
LH Blend #1 = 50% stevia, 50% monk fruit; LH Blend #2 = 40% stevia, 40% monk fruit, 20% allulose; LH Blend #3 = 60% monk fruit, 40% allulose; LH Blend #4 = 75% stevia, 25% monk fruit
Bolded percentages for JAR question attributes received significant penalty ($p<0.05$)

Table 17. Child consumer acceptance scores for 1% milkfat chocolate milks with different sweeteners (n=128)

		Sucrose Control	LH Sucrose	LH Stevia	LH Blend #2	LH Blend #4
Color Liking		5.7a	5.5a	5.5a	5.7a	5.5a
Aroma Liking		5.3a	5.4a	5.3a	5.3a	5.1a
Overall Liking		5.7a	5.8a	5.7a	5.9a	6.0a
Chocolate JAR	Not Enough Chocolate	**35.2%a**	**28.1%a**	**27.3%a**	**32.0%a**	**20.3%a**
	Just about Right	54.7%a	62.5%a	60.9%a	55.5%a	68.8%a
	Too Much Chocolate	10.2%a	9.4%a	11.7%a	12.5%a	10.9%a
Sweetness JAR	Not Sweet Enough	**25.0%a**	19.5%a	**21.1%a**	**22.7%a**	14.1%a
	Just About Right	55.5%a	64.8%a	56.3%a	65.6%a	62.5%a
	Too Sweet	19.5%a	15.6%a	**22.7%a**	11.7%a	**23.4%a**
Purchase Intent		3.6a	3.7a	3.6a	3.8a	3.8a
Compared to School Milk		3.7a	3.9a	3.8a	3.9a	3.9a

Means within a row followed by a different letter represent significant differences ($p<0.05$)
Liking questions were scored on a 7-point hedonic scale where dislike extremely=1 and like extremely=7
JAR questions were scored on a 5-point scale where 1 or 2 = too little, 3 = just about right, and 4 or 5 = too much
Purchase Intent question was scored on a 5-point scale where 1-2=would not purchase, 3= maybe/maybe not purchase, and 4-5=would purchase. Statistical lettering was determined using Kruskal-Wallis testing
Compared to School Milk question was scored on a 5-point scale where 1-2= I like it less, 3= I like it the same, and 4-5=I like it more. Statistical lettering was determined using Kruskal-Wallis testing
LH Blend #2 = 40% stevia, 40% monk fruit, 20% allulose; LH Blend #4 = 75% stevia, 25% monk fruit.
LH: Lactose hydrolyzed
Bolded percentages for JAR question attributes received significant penalty ($p<0.05$

APPENDICES

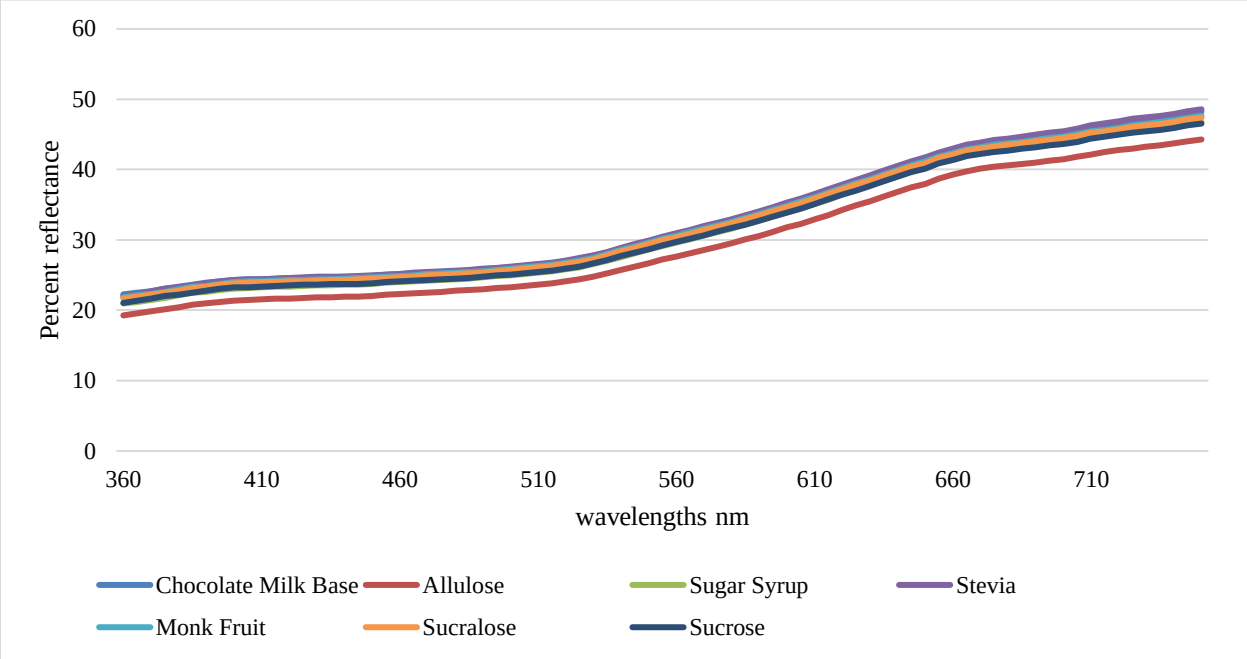
Appendix A. Cost of production of 1% milkfat chocolate milk with or without lactose hydrolysis and different sweeteners (per 100lb. chocolate milk batch)

	Sucrose Control	LH Sucrose	LH Stevia	LH Blend #1	LH Blend #2	LH Blend #3	LH Blend #4
Cost of sweetener ($)/100 lb. chocolate milk batch	1.72	3.02	2.50	4.04	5.13	7.14	3.27

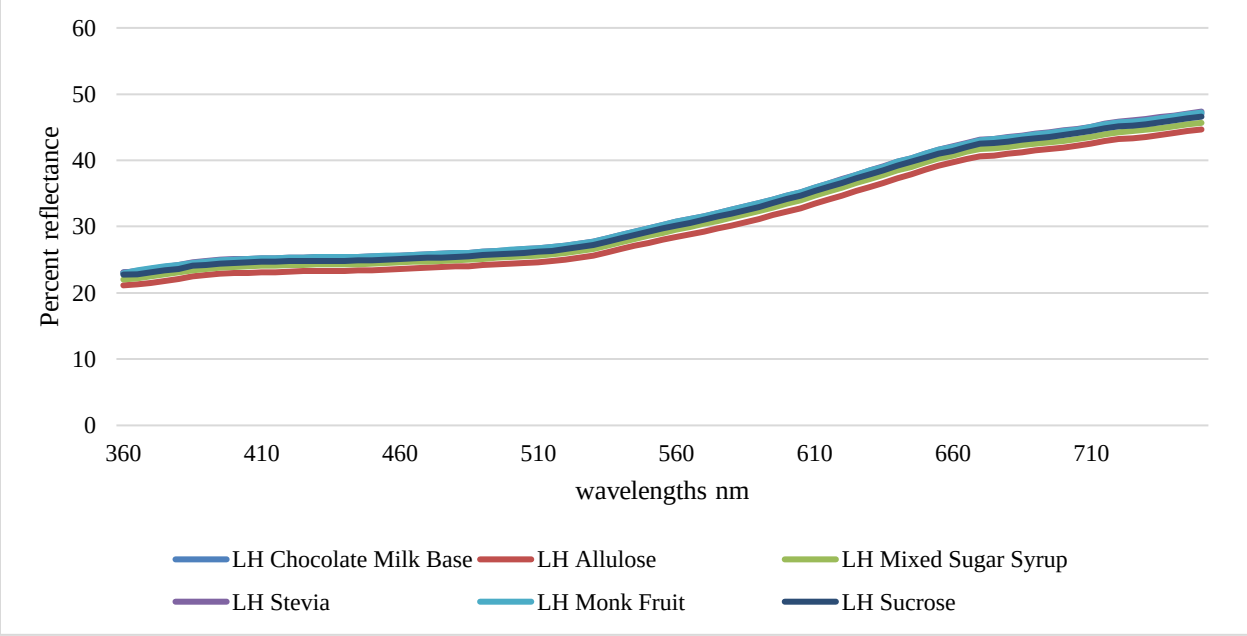
Cost of sweetener ($)/ 100 lb. chocolate milk batch represents the cost to produce 100 lb. of chocolate milk with the specific sweetener at the iso-sweet concentration of 3.69% w/w sucrose in 1% milkfat chocolate milk solution

LH Blend #1 = 50% stevia, 50% monk fruit; LH Blend #2 = 40% stevia, 40% monk fruit, 20% allulose; LH Blend #3 = 60% monk fruit, 40% allulose; LH Blend #4 = 75% stevia, 25% monk fruit

Appendix B. Reflectance curves for 1% milkfat chocolate milks in experiment 1

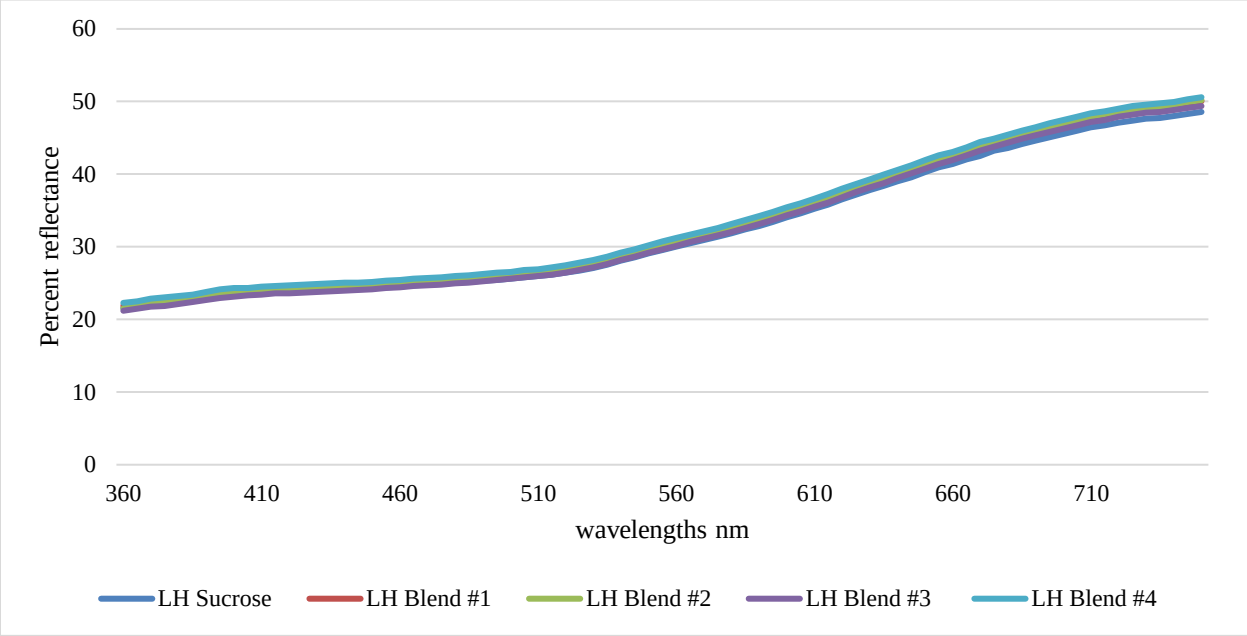


Appendix C. Reflectance curves for 1% milkfat lactose hydrolyzed (LH) chocolate milks in experiment 2. LH:



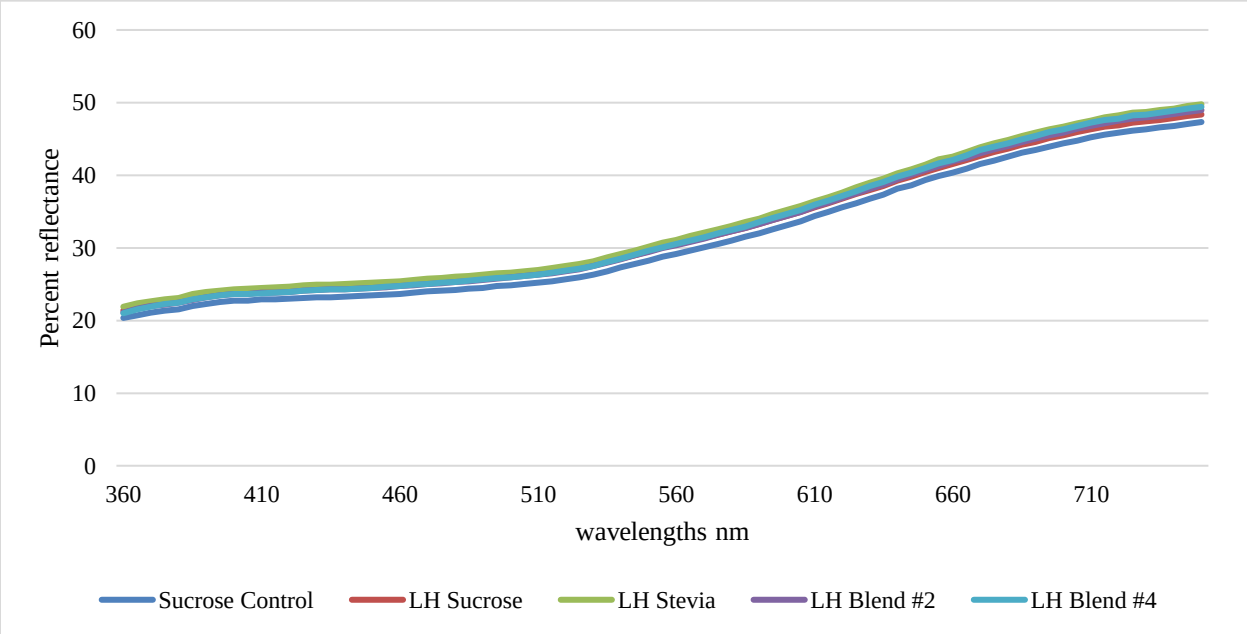
Lactose hydrolyzed.

Appendix D. Reflectance curves for 1% milkfat lactose hydrolyzed (LH) chocolate milks in experiment 3.



LH Blend #1 = 50% stevia, 50% monk fruit; LH Blend #2 = 40% stevia, 40% monk fruit, 20% allulose; LH Blend #3 = 60% monk fruit, 40% allulose; LH Blend #4 = 75% stevia, 25% monk fruit. LH: Lactose hydrolyzed.

Appendix E. Reflectance curves of 1% milkfat chocolate milk with or without lactose hydrolysis in children's acceptance test.



LH Blend #2 = 40% stevia, 40% monk fruit, 20% allulose; LH Blend #4 = 75% stevia, 25% monk fruit. LH: Lactose hydrolyzed.

Appendix F. Total solids and fat content of 1% milkfat chocolate milks in experiment 1

	Chocolate Milk Base	Sucrose	Stevia	Allulose	Monk Fruit	Mixed Sugar Syrup	Sucralose
Total Solids (%)	10.7 ± 0.02	14.2 ± 0.03	10.8 ± 0.04	16.9 ± 0.03	10.8 ± 0.02	14.6 ± 0.04	10.8 ± 0.01

Total solids content (%) of chocolate milks was measured in duplicate.
LH: Lactose hydrolyzed.

Appendix G. Total solids and fat content of 1% milkfat lactose hydrolyzed (LH) chocolate milks in experiment 2

	LH Chocolate Milk Base	LH Sucrose	LH Allulose	LH Monk Fruit	LH Mixed Sugar Syrup	LH Stevia
Total Solids (%)	10.8 ± 0.04	12.9 ± 0.03	14.9 ± 0.01	10.7 ± 0.02	13.1 ± 0.01	10.7 ± 0.02

Total solids content (%) of chocolate milks was measured in duplicate.
LH: Lactose hydrolyzed.

Appendix H. Total solids and fat content of 1% milkfat lactose hydrolyzed (LH) chocolate milks in experiment 3

	LH Chocolate Milk Base	LH Sucrose	LH Blend #1	LH Blend #2	LH Blend #3	LH Blend #4
Total Solids (%)	10.6 ± 0.02	13.0 ± 0.03	10.6 ± 0.01	11.4 ± 0.01	12.2 ± 0.03	10.6 ± 0.02

Total solids content (%) of chocolate milks was measured in duplicate.
LH: Lactose hydrolyzed.
LH Blend #1 = 50% stevia, 50% monk fruit; LH Blend #2 = 40% stevia, 40% monk fruit, 20% allulose; LH Blend #3 = 60% monk fruit, 40% allulose; LH Blend #4 = 75% stevia, 25% monk fruit.

Appendix I. Total solids and fat content of 1% milkfat chocolate milk with or without lactose hydrolysis in children's acceptance test

	Chocolate Milk Base	LH Chocolate Milk Base	Sucrose Control	LH Sucrose	LH Stevia	LH Blend #2	LH Blend #4
Total Solids (%)	11.0 ± 0.01	10.9 ± 0.03	14.3 ± 0.01	13.1 ± 0.05	10.9 ± 0.04	11.7 ± 0.02	10.9 ± 0.01

Total solids content (%) of chocolate milks was measured in duplicate.
LH: Lactose hydrolyzed.
LH Blend #2 = 40% stevia, 40% monk fruit, 20% allulose; LH Blend #4 = 75% stevia, 25% monk fruit.

Appendix J. Demographic information and consumer consumption characteristics of young adult acceptance test of 1% milkfat chocolate milk in experiment 1 (n=122)

Question	Criteria	%
Please indicate your gender.	Male	31.1%
	Female	68.0%
	Non-binary	0.8%
	Other	0.0%
Which age group do you belong?	18-24 years old	36.1%
	25-34 years old	14.8%
	35-44 years old	7.4%
	45-54 years old	7.4%
	56-64 years old	0.0%
	65 years old and over	0.0%
Please indicate your ethnicity.	Asian/Pacific Islander	17.2%
	Black/African American	4.9%
	Hispanic/Latino	6.6%
	White/Caucasian	68.9%
	Other (please specify)	2.5%
How often do you consume chocolate milk?	Daily	38.5%
	Weekly	48.4%
	2-3 times a month	7.4%
	Monthly	4.9%
	3-4 times year	0.8%
	Once or twice a year	0.0%
	1 don't consume milk	0.0%
How often do you consume chocolate milk?	Daily	5.7%
	Weekly	38.5%
	2-3 times a month	23.8%
	Monthly	27.9%
	3-4 times year	4.1%
	Once or twice a year	0.0%
	1 don't consume milk	0.0%
Which fat content milk do you consume most often?	Skim or nonfat	6.6%
	Low fat or 1%	25.4%
	Reduced fat or 2%	38.5%
	Whole or regular	29.5%

Data represents 122 consumers

Demographics may vary slightly from requested due to show rates

1. Comments for “Other” included “Puerto Rican” (n=1), “Portuguese” (n=1), and “Black and white” (n=1)

Appendix K. Demographic information and consumer consumption characteristics of young adult acceptance test of 1% milkfat lactose hydrolyzed chocolate milk in experiment 2 (n=160)

Question	Criteria	%
Please indicate your gender.	Male	31.1%
	Female	68.0%
	Non-binary	0.8%
	Other	0.0%
Which age group do you belong?	18-24 years old	36.9%
	25-34 years old	38.8%
	35-44 years old	8.8%
	45-54 years old	10.6%
	56-64 years old	5.0%
	65 years old and over	0.0%
Please indicate your ethnicity.	Asian/Pacific Islander	18.1%
	Black/African American	4.4%
	Hispanic/Latino	11.3%
	White/Caucasian	64.4%
	Other (please specify)[1]	1.9%
How often do you consume chocolate milk?	Daily	38.1%
	Weekly	38.1%
	2-3 times a month	15.0%
	Monthly	7.5%
	3-4 times year	1.3%
	Once or twice a year	0.0%
	1 don't consume milk	0.0%
How often do you consume chocolate milk?	Daily	11.9%
	Weekly	30.0%
	2-3 times a month	28.1%
	Monthly	26.3%
	3-4 times year	3.8%
	Once or twice a year	0.0%
	1 don't consume chocolate milk	0.0%
Which fat content milk do you consume most often?	Skim or nonfat	6.3%
	Low fat or 1%	28.1%
	Reduced fat or 2%	35.0%
	Whole or regular	30.6%

Data represents 160 consumers

Demographics may vary slightly from requested due to show rates

1. Comments for "Other" included "Amerasian" (n=1), "Portuguese" (n=1), and "Middle Eastern/North African" (n=1)

Appendix L. Demographic information and consumer consumption characteristics of young adult acceptance test of 1% milkfat lactose hydrolyzed chocolate milk in experiment 3 (n=131)

Question	Criteria	%
Please indicate your gender.	Male	31.3%
	Female	66.4%
	Non-binary	2.3%
	Other	0.0%
Which age group do you belong?	18-24 years old	51.1%
	25-34 years old	29.0%
	35-44 years old	6.9%
	45-54 years old	5.3%
	56-64 years old	7.6%
	65 years old and over	0.0%
Please indicate your ethnicity.	Asian/Pacific Islander	29.8%
	Black/African American	7.6%
	Hispanic/Latino	5.3%
	White/Caucasian	55.7%
	Other (please specify)[1]	1.5%
How often do you consume chocolate milk?	Daily	36.6%
	Weekly	35.1%
	2-3 times a month	16.0%
	Monthly	10.7%
	3-4 times year	1.5%
	Once or twice a year	0.0%
	1 don't consume milk	0.0%
How often do you consume chocolate milk?	Daily	10.7%
	Weekly	29.8%
	2-3 times a month	26.0%
	Monthly	28.2%
	3-4 times year	5.3%
	Once or twice a year	0.0%
	1 don't consume chocolate milk	0.0%
Which fat content milk do you consume most often?	Skim or nonfat	5.3%
	Low fat or 1%	32.8%
	Reduced fat or 2%	33.6%
	Whole or regular	28.2%

Data represents 131 consumers

Demographics may vary slightly from requested due to show rates

1. Comments for "Other" included "Asian/white" (n=1), "black/white" (n=1)

Appendix M. Demographic information and consumer consumption characteristics of children's acceptance test of 1% milkfat chocolate milk with different sweeteners in experiment 3 (n=128)

Question	Criteria	%
What is the gender or your child?	Male	53.9%
	Female	46.1%
What is the age of your child?	Under 2 years old	0.0%
	3-5 years old	0.0%
	6-7 years old	0.0%
	8-9 years old	31.3%
	10-13 years old	68.8%
	14-17 years old	0.0%
	18+ years old	0.0%
What type of school does your child attend?	Public	89.1%
	Private	10.9%
	Homeschooled	0.0%
How often does your child typically purchase lunch at school?	Daily	29.7%
	4-6 days per week	10.2%
	2-3 days per week	21.9%
	Once a week	20.3%
	2-3 times per month	18.0%
	Once a month or less	0.0%
	Never	0.0%
Which beverages does your child consume? Select all that apply.[1]	Juice	93.0%
	Soft Drinks	75.0%
	Milk	100.0%
	Flavored Water	78.1%
How often does you- child typically purchase lunch at school?	Daily	35.2%
	4-6 days per week	15.6%
	2-3 days per week	13.3%
	Once a week	20.3%
	2-3 times per month	15.6%
	Once a month or less	0.0%
	Never	0.0%
What types of milk does your child drink? Select all that apply.[1]	Plain milk, fat-free	40.6%
	Plain milk, low-fat	79.7%
	Plain milk, whole	60.2%
	Flavored milk, fat-free	52.3%
	Flavored milk, low-fat	98.4%
	Flavored milk, whole	60.9%

Appendix M. Demographic information and consumer consumption characteristics of children's acceptance test of 1% milkfat chocolate milk with different sweeteners in experiment 3 (n=128) (continued)

What type of milk does your child typically drink most often?	Plain milk, fat-free	0.8%
	Plain milk, low-fat	33.6%
	Plain milk, whole	10.2%
	Flavored milk, fat-free	8.6%
	Flavored milk, low-fat	35.9%
	Flavored milk, whole	10.9%

Data represents 128 child consumers
Demographics may vary slightly from requested due to show rates
Total percentage exceeds 100% as consumers were able to select any responses that applied.

Appendix N. Mean drops of penalty analysis of young adult acceptance test of 1% milkfat chocolate milk in experiment 1 (n=122)

Question	Attribute	Sucrose Control	Allulose	Mixed Sugar Syrup	Monk Fruit	Stevia	Sucralose
Color JAR	Too Light	-	0.9	0.9	1.0	1.1	0.9
	Just About Right	-	-	-	-	-	-
	Too Dark	-	-	-	-	-	-
Aroma JAR	Too Weak	0.8	-	0.7	-	-	-
	Just About Right	-	-	-	-	-	-
	Too Strong	-	-	-	-	-	-
Overall Flavor JAR	Too Weak	2.0	2.1	1.8	1.5	1.5	1.3
	Just About Right	-	-	-	-	-	-
	Too Strong	-	-	-	2.5	-	-
Sweetness JAR	Not Sweet Enough	-	1.9	1.3	1.3	-	-
	Just About Right	-	-	-	-	-	-
	Too Sweet	-	-	-	2.4	1.9	1.5
Chocolate Flavor JAR	Too Weak	1.7	1.6	1.4	1.7	1.6	1.2
	Just About Right	-	-	-	-	-	-
	Too Strong	-	-	-	-	-	-
Mouthfeel/ Thickness JAR	Too Thin	-	1.5	1.8	-	1.1	1.2
	Just About Right	-	-	-	-	-	-
	Too Thick	-	-	-	-	-	-
Aftertaste JAR	Too Weak	-	-	-	-	-	-
	Just About Right	-	-	-	-	-	-
	Too Strong	1.8	2.0	-	1.7	1.7	1.3

Mean drop of penalty analysis determines if respondents' "Just About Right" ratings for a specific attribute are associated with a drop in Overall Liking rating.

Appendix O. Means drops of penalty analysis of young adult acceptance test of 1% milkfat lactose hydrolyzed chocolate milk in experiment 2 (n=160)

Question	Attribute	Sucrose Control	LH Sucrose	LH Allulose	LH Mixed Sugar Syrup	LH Monk Fruit	LH Stevia
Color JAR	Too Light	1.0	0.8	1.1	0.7	-	-
	Just About Right	-	-	-	-	-	-
	Too Dark	-	-	-	-	-	-
Aroma JAR	Too Weak	0.7	0.6	0.3	1.4	0.5	0.4
	Just About Right	-	-	-	-	-	-
	Too Strong	-	-	-	-	-	-
Overall Flavor JAR	Too Weak	1.8	1.8	1.9	1.9	1.6	1.9
	Just About Right	-	-	-	-	-	-
	Too Strong	-	-	-	-	-	-
Sweetness JAR	Not Sweet Enough	1.9	1.2	2.2	1.7	-	-
	Just About Right	-	-	-	-	-	-
	Too Sweet	-	-	-	-	1.5	2.0
Chocolate Flavor JAR	Too Weak	1.5	1.5	2.0	1.7	1.3	1.5
	Just About Right	-	-	-	-	-	-
	Too Strong	-	-	-	-	-	-
Mouthfeel/Thickness JAR	Too Thin	0.7	1.2	1.5	-	0.9	-
	Just About Right	-	-	-	-	-	-
	Too Thick	-	-	-	-	-	-
Aftertaste JAR	Too Weak	-	-	-	-	-	-
	Just About Right	-	-	-	-	-	-
	Too Strong	-	-	1.4	1.1	1.8	1.4

LH: Lactose hydrolyzed.

Mean drop of penalty analysis determines if respondents' "Just About Right" ratings for a specific attribute are associated with a drop in Overall Liking rating.

Appendix P. Mean drops of penalty analysis of young adult acceptance test of 1% milkfat lactose hydrolyzed chocolate milk in experiment 3 (n=131)

Question	Attribute	Sucrose Control	LH Sugar	LH Blend #1	LH Blend #2	LH Blend #3	LH Blend #4
Color JAR	Too Light	-	0.8	-	0.8	0.6	-
	Just About Right	-	-	-	-	-	-
	Too Dark	-	-	-	-	-	-
Aroma JAR	Too Weak	-	0.7	-	1.1	0.6	0.7
	Just About Right	-	-	-	-	-	-
	Too Strong	-	-	-	-	-	-
Overall Flavor JAR	Too Weak	1.2	1.6	2.0	1.8	2.0	1.6
	Just About Right	-	-	-	-	-	-
	Too Strong	-	-	-	-	-	-
Sweetness JAR	Not Sweet Enough	-	1.6	-	-	1.5	-
	Just About Right	-	-	-	-	-	-
	Too Sweet	-	-	1.7	0.9	-	1.8
Chocolate Flavor JAR	Too Weak	0.9	1.4	1.2	1.1	1.5	1.5
	Just About Right	-	-	-	-	-	-
	Too Strong	-	-	-	-	-	-
Mouthfeel/Thickness JAR	Too Thin	-	0.9	-	1.4	1.2	-
	Just About Right	-	-	-	-	-	-
	Too Thick	-	-	-	-	-	-
Aftertaste JAR	Too Weak	-	-	-	-	-	-
	Just About Right	-	-	-	-	-	-
	Too Strong	-	-	2.5	1.4	1.5	2.1

LH: Lactose hydrolyzed

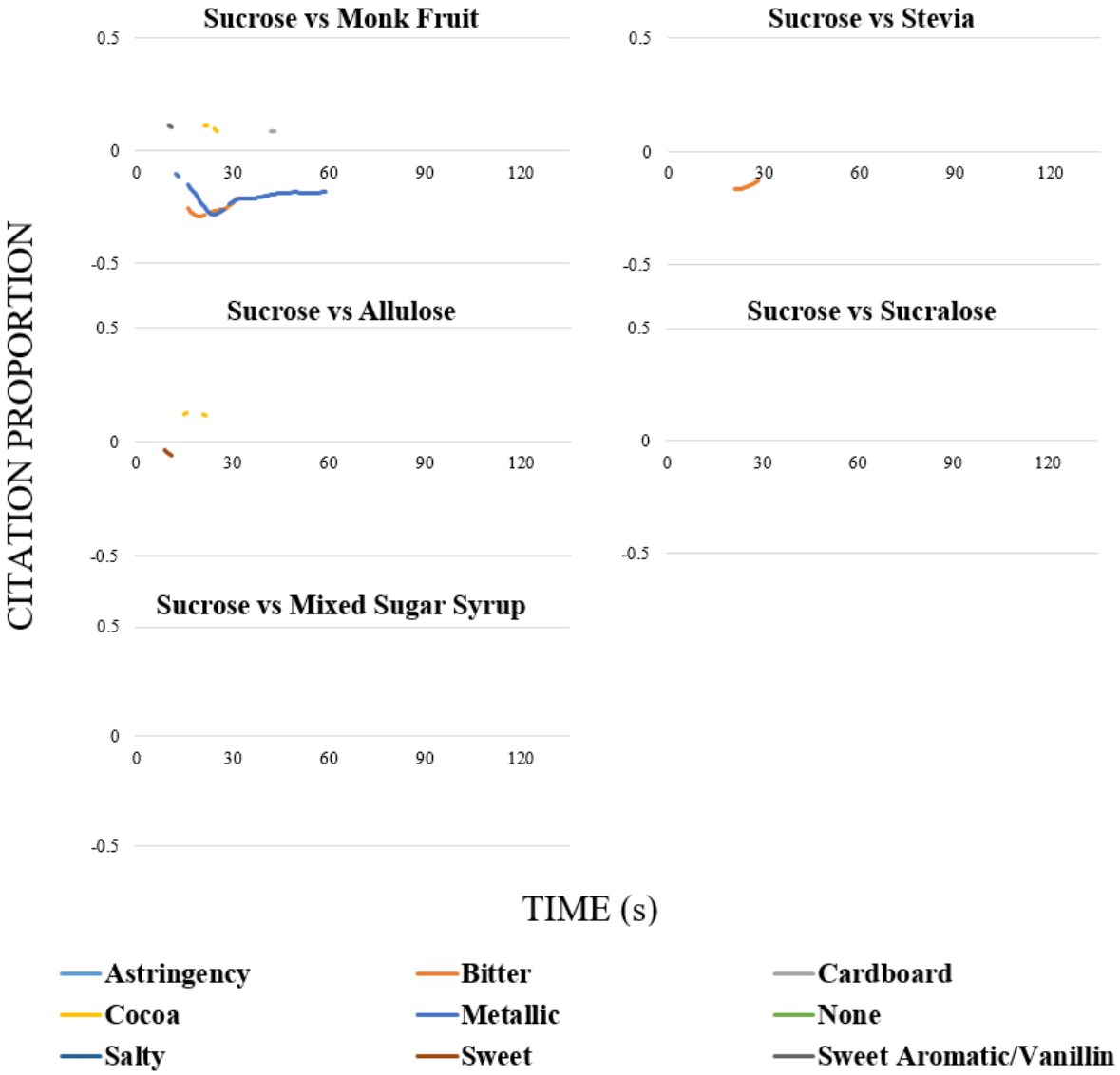
Mean drop of penalty analysis determines if respondents' "Just About Right" ratings for a specific attribute are associated with a drop in Overall Liking rating.

Appendix Q. Means drop for penalty analysis for of children's acceptance test of 1% milkfat chocolate milk with different sweeteners in experiment 3 (n=128)

Question	Attribute	Sucrose Control	LH Sucrose	LH Stevia	LH Blend #2	LH Blend #4
Chocolate JAR	Not Enough Chocolate	2.0	1.3	1.4	1.5	0.9
	Just About Right	-	-	-	-	-
	Too Much Chocolate	-	-	-	-	-
Sweetness JAR	Not Sweet Enough	1.7	-	1.3	1.3	-
	Just About Right	-	-	-	-	-
	Too Sweet	-	-	1.4	-	0.7

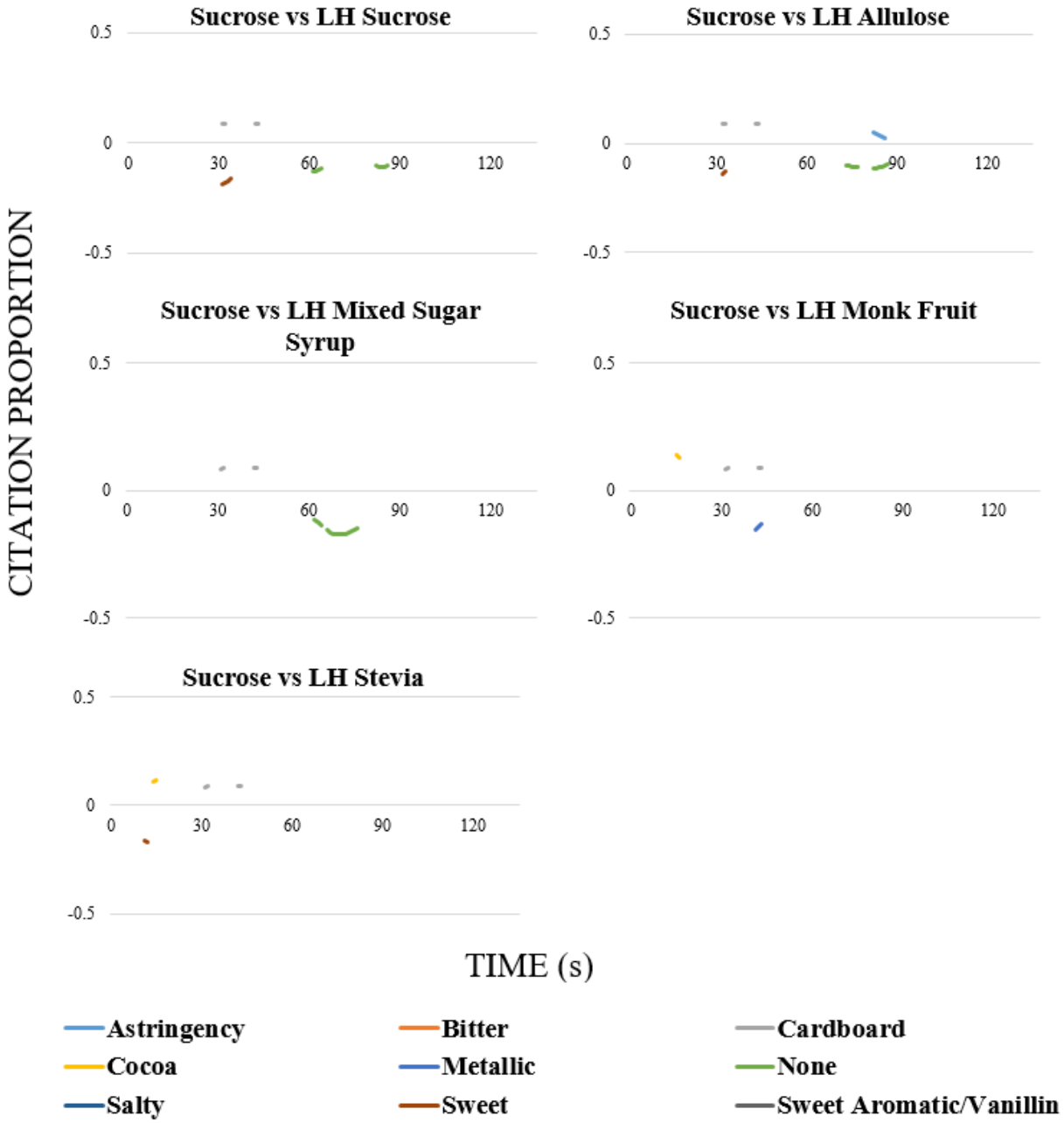
LH: Lactose hydrolyzed.
Mean drop of penalty analysis determines if respondents' "Just About Right" ratings for a specific attribute are associated with a drop in Overall Liking rating.

Appendix R. Difference ($p<0.05$) in temporal dominance of sensations (TDS) curves for nutritive and nonnutritive sweeteners in chocolate milk compared to the sucrose control chocolate milk.



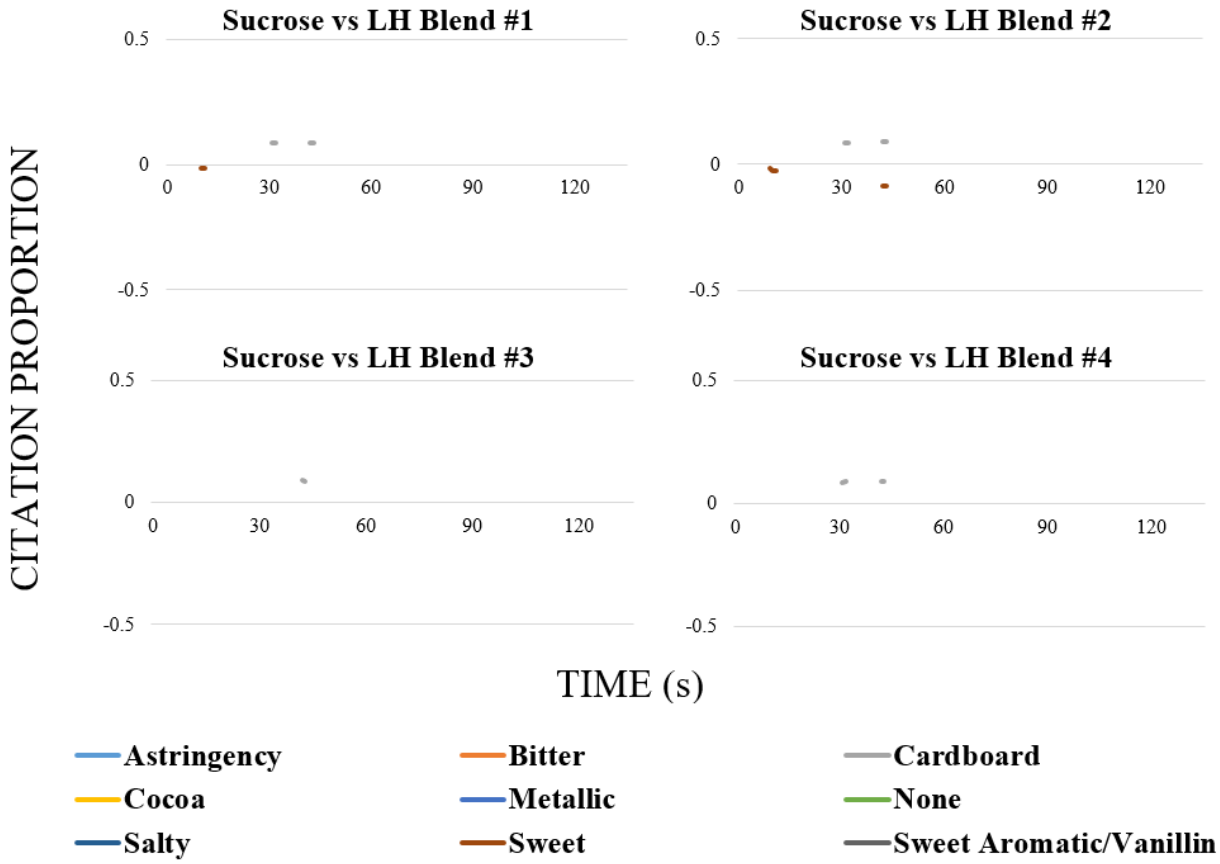
For all graphs, samples evaluation in mouth conducted at 0 to 15 s, aftertaste evaluation conducted at 20 to 135 s. For all graphs, difference in citation proportion for sucrose sweetened chocolate milk (positive citation proportion) compared to nutritive/nonnutritive sweetener chocolate milks (negative citation proportion).

Appendix S. Difference ($p<0.05$) in temporal dominance of sensations (TDS) curves for nutritive and nonnutritive sweeteners in lactose hydrolyzed chocolate milk compared the sucrose control chocolate milk.



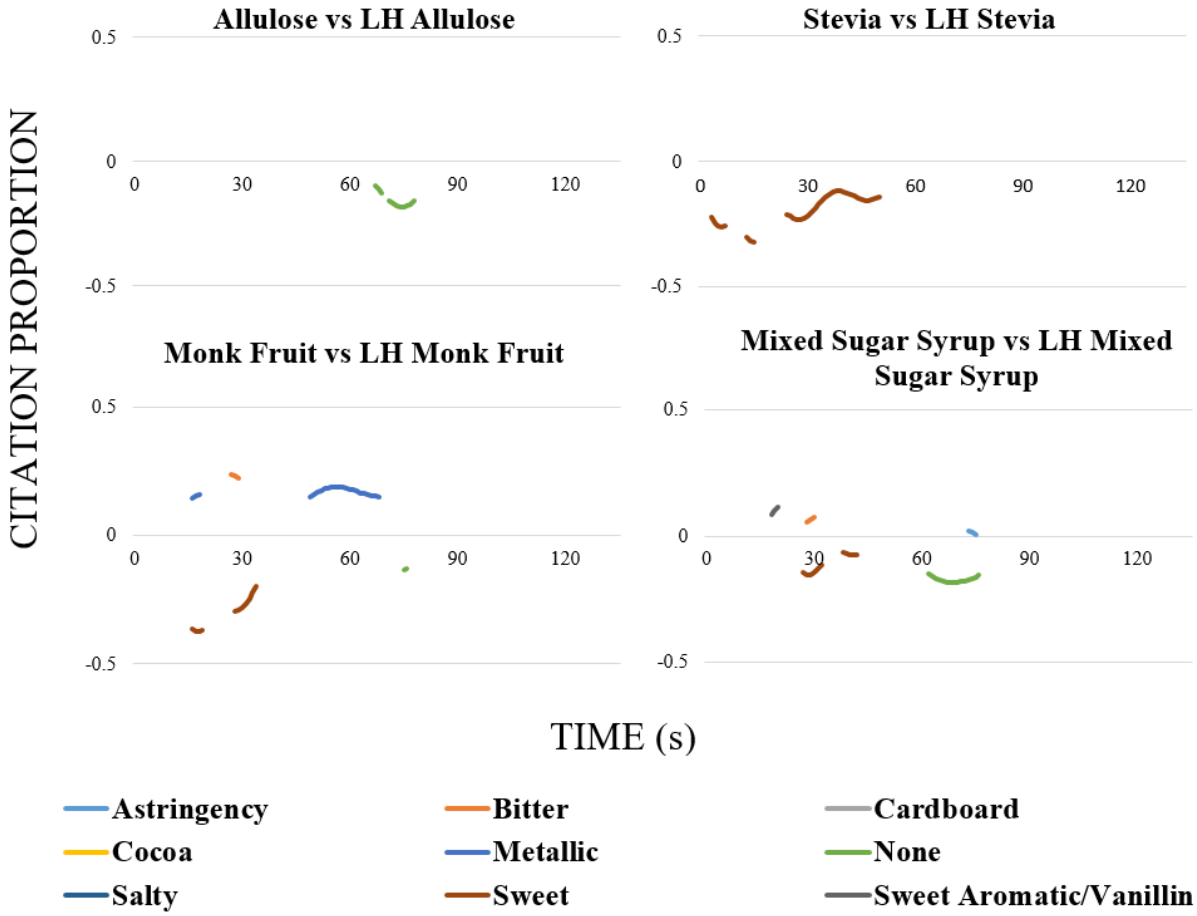
For all graphs, samples evaluation in mouth conducted at 0 to 15 s, aftertaste evaluation conducted at 20 to 135 s. For all graphs, difference in citation proportion for sucrose sweetened chocolate milk (positive citation proportion) compared to nutritive/nonnutritive sweetener lactose hydrolyzed chocolate milks (negative citation proportion).

Appendix T. Difference ($p<0.05$) in temporal dominance of sensations (TDS) curves for sweeteners blends in lactose hydrolyzed chocolate milk compared the sucrose control chocolate milk.



For all graphs, samples evaluation in mouth conducted at 0 to 15 s, aftertaste evaluation conducted at 20 to 135 s. For all graphs, difference in citation proportion for sucrose sweetened chocolate milk (positive citation proportion) compared sweetener blend lactose hydrolyzed chocolate milks (negative citation proportion).

Appendix U. Difference ($p<0.05$) in temporal dominance of sensations (TDS) curves for nutritive and nonnutritive sweeteners in chocolate milk and lactose-hydrolyzed chocolate milk.



For all graphs, samples evaluation in mouth conducted at 0 to 15 s, aftertaste evaluation conducted at 20 to 135 s. For all graphs, difference in citation proportion for nutritive and nonnutritive sweeteners in chocolate milk (positive citation proportion) compared to lactose-hydrolyzed milk (negative citation proportion).