ABSTRACT

HARWOOD, WILLIAM SPENCER. Investigation of Consumer Perceptions and Sensory Qualities Related to Fluid Milk and Dairy-Based Beverages (Under the direction of Dr. MaryAnne Drake).

Understanding consumer choice requires a holistic understanding of consumer beliefs, sensory drivers of preference, inherent product features, and an understanding of the tools by which these insights are garnered. The objective of this dissertation was threefold: to investigate and define consumer preferences for fluid milk, to address practical issues related to milk processing and storage, and, lastly, to propose novel methods for understanding milk and dairy beverages. Four different studies were conducted: validation of fluid milk consumer segments using quantitative multivariate analysis, the role of heat treatment on light oxidation of fluid milk, the influence of automatic associations on preference for milk type, and the application of temporal penalty analysis for the optimization of sugar reduction in protein beverages

In the first study, the validity of fluid milk consumer segments derived from a large-scale conjoint analysis survey was assessed using qualitative sensory techniques- an approach called qualitative multivariate analysis (QMA). Conjoint analysis results suggested fluid milk consumer segmentation was primarily based upon price sensitivity and milk type (conventional, organic, pasture-raised, or local). Segment-specific differences related to price and milk type were verified with home usage testing and focus group evaluations; however, these belief systems and preference patterns were largely based on quality perception, alignment with personal beliefs, and intended use of product (ex: as a beverage, as an ingredient, as a base for cereal, etc.). Within the milk industry, the joint approach of large-scale consumer surveys and qualitative assessments may provide valuable consumer-relevant context, thus serving as a valuable resource for marketing, education, and new product development campaigns.

In the second study, the effect of thermal processing on light oxidized flavor (LOF) development in fluid milk was investigated. Milks processed using high temperature short time (HTST) or direct steam injection ultrapasteurization (DSI-UP) were compared using consumer threshold testing, volatile flavor analysis, and trained panel sensory profiling techniques. Trained panel and consumer threshold results proved that the onset of LOF was significantly delayed in DSI-UP milk compared to HTST milk. Inhibition of LOF development in DSI-UP milk was largely attributed to the formation of volatile sulfur compounds, which likely conferred antioxidant and sensory masking effects.

In the third study, consumer preferences for different milk types (conventional, organic, local, or pasture-raised) were assessed from both an implicit and explicit perspective. Implicit beliefs related to milk type were assessed in pairwise comparisons using computer-based implicit association tests (IATs). In addition, consumer preferences for different milk types were assessed in blinded and primed preference test evaluations. Results indicated that milk consumers hold strong negative implicit biases against conventional fluid milks, and that beliefs related to non-conventional milk types were largely conflated. Furthermore, this study showed clear evidence that fluid milk consumers were willing to override their sensory preferences when milk type was disclosed to them to express alignment with their stated beliefs.

In the fourth study, a novel analysis method for temporal data was proposed in order to optimize formulation of protein beverages with natural nonnutritive sweeteners. Temporal check-all-that-apply (TCATA) and temporal liking (TL) data streams from untrained consumers were combined using a penalty analysis approach at each second of evaluation to understand the temporal sensory features that affect consumer acceptance. Formulations using single natural nonnutritive sweeteners (stevia or monk fruit) were associated with extended periods of sensory

penalties related to metallic flavor and bitter taste, which were not present in the sucrose-sweetened formulation. These temporal sensory concerns were largely mitigated in natural nonnutritive sweetener blends, as evidenced by reduced penalties. Overall, temporal penalty analysis (TPA) proved to be an effective and measurable approach to assessing temporal sensory data from a consumer-relevant perspective.

Investigation of Consumer Perceptions and Sensory Qualities Related to Fluid Milk and Dairy-Based Beverages

by
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A dissertation submitted to the Graduate Faculty of
North Carolina State University
in partial fulfillment of the
requirements for the degree of
Doctor of Philosophy

Food Science

Raleigh, North Carolina

2020

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DEDICATION

This dissertation is dedicated to my wife, Kristy. You have been a shining example of love and support throughout this process and I have been so blessed to have you. I truly could not have achieved any of this without you by my side.

BIOGRAPHY

William Spencer Harwood was born on March 15th, 1993 to Bill and Stephanie Harwood. Will grew up in West Palm Beach, Florida, where he attended high school. Following high school, Will attended the University of Florida, where he graduated in 2015 with a B.S. in Food Science. After graduating college, Will married his high school sweetheart, Kristy, on July 12th, 2015. Following the wedding, the newlywed couple moved to Raleigh, NC where Will began pursuing a M.S. degree in Food Science and North Carolina State University under the direction of Dr. MaryAnne Drake. Will completed his M.S. degree at NC State in 2018 and decided to continue in the lab to pursue his PhD after. Away from school, Will enjoys basketball, volleyball, cooking, and walks with his dog, Bear.

ACKNOWLEDGMENTS

I’ve been lucky to have so many people by my side over the last few years. Thank you to my parents for your continued love and support, and for teaching me the value of perseverance and hard work. Thank you to the other MAD Lab members for all of your help along the way. Earning a degree in this lab is no easy task, but having a team that is full of intelligent, hardworking, and selfless individuals has made it far more enjoyable. I would like to thank my committee members, Dr. Hanson, Dr. Fogleman, Dr. Stevenson, and Dr. Poole, for their insight and guidance during my research. I would also like to give a special thanks to Dr. Drake. Dr. Drake, thank you for taking a chance on me and for being an incredible mentor these last few years. I can’t thank you enough for the time and effort you’ve put into helping me and making me the professional I am today.

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CHAPTER 1: The Impact of Processing on the Sensory Properties of Milk and Dairy Products

The Impact of Processing on the Sensory Properties of Milk and Dairy Products

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The content of this chapter was submitted for publication in:

Encyclopedia of Dairy Sciences, 3rd edition- Manufacture of Milk and Whey Products

INTRODUCTION

Fluid milk and dairy products are enjoyed throughout the world. The wide array of flavors and formats that make up this category can meet the needs of everyone from preschoolers to elite athletes. Furthermore, dairy products are unique vehicles for nutritional supplementation and functionality/product performance. The ubiquity and diversity of milk and dairy products can be attributed, at least in part, to the multiple processes that are used to prepare these items. Processing techniques applied within the dairy industry ensure that products reach consumers at the intersection of quality and convenience. For fluid milk and its derivatives, processing steps may be taken for various reasons. Thermal processing techniques, such as those made famous by Louis Pasteur, may be employed to reduce bacterial load and eliminate pathogens, thus providing safety and extended shelf life. Alternatively, spray drying of liquid whey or milk proteins offers advantages related to ease of transport in addition to lowered spoilage risk.

While the practical advantages achieved through processing and packaging may be clear, flavor and texture evolution resulting from such processes are not as obvious. However, an understanding of the relationship between processing relationship and the sensory profile of milk, whey products, and other dairy foods is essential, as consumer sensory acceptance often supersedes extrinsic factors (Kim et al., 2013; Parker et al., 2018; Hoque et al., 2018). Furthermore, flavor changes resulting from different processes may dictate the downstream approach of using certain milk derivatives as ingredients. This chapter will investigate the relationship between fluid milk and dairy products and their associated processes in the context of sensory profile. Knowledge of these relationships may serve as a powerful resource for manufacturers and product development initiatives within the dairy industry.

FLUID MILK PRE-PROCESSING

In order to fully investigate the sensory-related effects contributed by processing, it is important to understand the sensory profile of fluid milk and its constituents pre-processing. In general, fresh fluid raw milk is characterized by a minimal flavor and aroma profile, with sweet aromatic and fresh milkfat notes being the dominant sensory features. Grassy and silage/feed aromas and flavors will be evident depending on the feed source of the cow (Croissant et al., 2007). Because the sensory profile of fluid milk is relatively low-impact and ideally free from off-flavors, analysis of fresh fluid milk flavor and quality has conventionally been addressed through defect identification techniques. Defect monitoring for fluid milk and dairy products has traditionally been approached using quality judging techniques that use a scorecard system to identify, classify, and rate common sensory defects. These defects were initially documented in raw whole milk then high temperature short time (HTST) pasteurized milk and then most recently 2% fat HTST milk. The purpose of defect judging was to evaluate gross quality prior to product release to the consumer supply chain. This approach is still successfully utilized today as an appropriate quality assurance tool. Modern research and market studies related to milk flavor have employed the use of trained panel descriptive analysis techniques and other objective and subjective sensory measures to provide more appropriate guidance for research or consumer marketing objectives (Schiano et al., 2017). An overview of the array of tests for specific sensory objectives that can be applied to milk and dairy foods can be found in Figure 1. The reader is also encouraged to explore comprehensive reviews of sensory methodologies employed in the dairy industry elsewhere (Drake, 2007; Drake, 2008; Schiano et al., 2017).

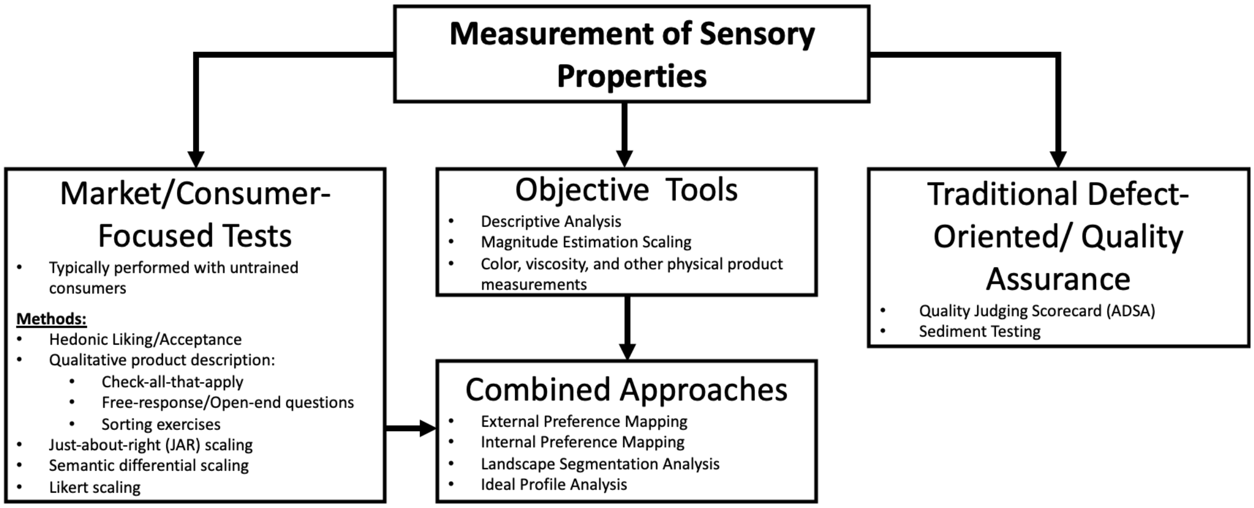

Figure 1. Overview of sensory approaches and associated methods
Adapted from Drake, 2007; Drake, 2008; Thompson et al., 2004; Meilgaard et al., 2006; Schiano et al., 2017

Non-Processing Flavor Contributors

Previous studies related to milk flavor and quality have identified and described an exhaustive collection of off-flavors (Table 1). Many of the documented deviations in flavor quality can be ascribed to the following categories: light oxidation, enzymatic activity (lipolysis, proteolysis), autooxidation, feed, and microbial spoilage. These off flavors are not directly the effects of processing but are reviewed briefly as they can occur in raw and pasteurized milk and may persist following processing.

Table 1. Common fluid milk off-flavors and their sources

Flavor Defect	Description	Training Reference
Acid	Sour off-flavor due to acid-producing organisms such as Lactococcus lactis subsp. Cremoris	Add 6-7mL of a 10% lactic acid solution to ~600 mL milk
Astringent*	Puckering sensation on tongue and lining of the mouth	n/a
Cowy/barny/unclean*	Distinct cow's breath-like odor and unpleasant, medicinal, or chemical aftertaste, suggestive of a poorly maintained barn	n/a
Bitter	Persistent bitter taste detected at the base of the tongue, commonly caused by specific weeds consumed as part of roughage by cows or by proteolysis of milk proteins by microorganisms (especially psychrotrophic bacteria)	Add 2-2.5 mL of a 0.1% quinine sulfate solution to ~600 mL milk
Cooked	Sulfurous, heated, caramelized, or scorched flavors	Heat a working quantity of milk in a vessel to 80˚C and hold for 1 min
Feed	Aromatic taints resulting from cows consuming some feeds within a critical time frame before milking. Have a characteristic "cleanliness" note and a mild aftertaste which disappears quickly.	Add 4-7 mL of a prepared "tea" (brew alfalfa in water) to 600 mL milk
Fermented/Fruity	May resemble the odor of sauerkraut, vinegar, pineapple, apples, or other fruits, commonly caused by the growth of microorganisms.	Mix 6 parts pineapple juice and one part vinegar. Add 3-4 mL mixture to 600 mL milk.
Flat	Simulated by adding water to a sample of milk and noticing the alteration of mouthfeel of the mixture	Add about 20% water to 2% fat milk
Foreign/chemical	Chemical flavor which may be caused by improper use of detergents, disinfectants, and sanitizers; exposure to gasoline or kerosene fumes; or contamination from insecticides or medicines	Add 2 mL of a 200 ppm chlorine solution to 600 mL milk immediately before presentation
Garlic/onion	Weedy, pungent odors and somewhat persistent aftertaste	Add 2 mL of a 1% garlic powder mix (in water) to the milk; infuse a clove or garlic for 2hrs then either decant the milk or retrieve the clove
Lacks Freshness	Stale, "chalky" flavor, lack of sweetness	Open a carton of milk and store in the refrigerator for ≥7 days; Use an unopened carton 1 wk beyond the pull date; Add 10-15g skim milk powder to 600 mL of milk

Table 1 (continued).

Malty	Suggestive of malt or Grape Nuts® cereal, generally caused by growth of Streptococcus lactis subsp. *Maltigenes*	Add 1g malt powder to 1 L warm milk; Add 15g Grape Nuts® to 100mL of milk and infuse for 20-30 min before adding aliquots to ~600mL milk
Oxidized - light	Bunt, burnt protein/feather-like, cabbage-like, medicinal, or chemical off-flavors resulting from light exposure	Transfer milk into a clear glass bottle and placed on a windowsill exposed to direct, bright sunlight for a duration proportional to the intensity of the defect
Oxidized - metal	Metallic, oily, cappy, cardboard, stale, tallowy, painty, or fishy off-flavor commonly induced by the catalytic action of certain metals. Characterized by a puckery mouthfeel.	Immerse a copper penny or wire in milk overnight; Add several drops 1% copper sulfate solution to 600mL milk and leave in a refrigerator for 24 hrs
Rancid	Baby burp, feta cheese, or butyric acid aromas formed as a result of lipid hydrolysis	Add 0.5g lipase powder to ~600mL milk, agitate and hold at 21˚C for 1hr;Add a few drops of dilute butyric acid solution to ~600mL milk
Salty	Commonly associated with milk from cows in advanced stages of lactation or with clinical mastitis, resulting in an increase of NaCl in the milk and a decrease of other mineral salts	Dissolve 0.25-0.5g table salt into 600mL milk.
Unclean	Offensive odor suggesting extreme staleness, mustiness, dirty socks, or foul stable air. May develop due to the action of certain psychrotrophic bacteria.	Combine rancid, fruity, and bitter milks; mix spoiled milk (≥7-10 days beyond sell by date) with fresh milk

Taken from Schiano et al. (2017)

Light Oxidation

Light-induced oxidation and off-flavor development within fluid milk and dairy products has been common knowledge in the dairy industry for several years (Schiano et al., 2017). Components of fluid milk including riboflavin (vitamin B_2) and naturally occurring tetrapyrroles are readily photosensitized, and subsequently act as catalysts in the oxidation of amino acids and unsaturated fatty acids when oxygen is present (Min and Boff, 2002). Riboflavin-mediated photooxidation of amino acids and lipids within milk and dairy products is associated with the development of volatile aldehydes and related compounds that contribute oxidized flavors described as cardboard, mushroom, or butterscotch (Brothersen et al., 2016; Harwood et al., 2020). While the off flavor and the specific sensory nature of the off flavor may depend on product features such as milkfat (skim milk is generally more susceptible to light oxidation and riboflavin loss and the flavor is more mushroom and cardboard compared to whole milk where the off flavor is more butterscotch and rubbery in nature) and applied thermal processes (cooked flavors may help mask light oxidized flavor), the onset of such flavors may be apparent in fluid milk in as little as 2 hours (Whited et al., 2002; Chapman et al., 2002; Harwood et al., 2020). Furthermore, the sensory effects associated with light oxidation have been shown to increase linearly with extended exposure time (Whited et al., 2002). Mitigation of light oxidized flavor development within milk and dairy products is typically approached through antioxidant addition (van Aardt et al., 2005; Hall et al., 2010) light-blocking package design (Moyssiadi et al., 2004; Mestadgh et al., 2005; Stancik et al., 2017), unit processes such as homogenization and thermal processing (Saidi and Warthesen, 1995; Harwood et al., 2020), or combinatorial approaches. Light oxidation and the associated light oxidized flavor (LOF) can occur in all dairy foods, not just fluid milk. The LOF flavor in other dairy foods may be due to the use of LOF milk or ingredients (milk powder made from LOF milk) or the LOF flavor may due to light exposure of the dairy food itself (yogurt and ice cream packaged in transparent glass or plastic packaging).

Enzymatic Activity

Fresh raw milk has a host of native enzymes designed to promote proper digestive health within suckling calves. However, during collection of milk for human consumption, the presence of these enzymes poses risk of deleterious sensory effects. Among the most common enzymes in milk that relate to sensory quality are alkaline phosphatase, lipase, proteinase (plasmin), and lactoperoxidase (Kelly and Fox, 2006). In general, enzymes native to fluid milk are inactivated following thermal processing; however, enzymatic activity pre-processing may confer significant off-flavors that survive processing. Lipolytic activity resulting from native milk lipase has been shown to cause hydrolytic rancidity (rancid off flavor) due to cleavage of ester bonds on triglycerides to release free fatty acids. The native milk lipase is in the serum phase of milk and the milkfat globule membrane surrounding the milkfat in raw milk protects it from lipase activity. However, if raw milk is mishandled (excessive agitation or pumping) and the milkfat globule membrane is broken, lipase can access milkfat and release of free fatty acids from triglycerides can occur. The characteristic short chain fatty acids in bovine milkfat, namely butyric acid, are flavor active and remain unchanged following heat treatment. In pasteurized milk, this off flavor is readily apparent to most consumers at amounts near 0.32 meq of FFA/kg of milk (Santos et al., 2003). Additionally, heat-stable proteolytic enzymes of native or bacterial origin can survive thermal processing and other unit operations which may subsequently result in undesirable gelation and/or off flavors during storage (Valero et al, 2001). Caution during pre-processing raw milk handling and attention to raw milk quality indicators such as somatic cell count as well as microbial counts and assessment of raw milk aroma and or flavor following heat treatment may dictate susceptibility to enzymatic off-flavor development post-processing (Ma et al., 2000).

Autooxidation

Autooxidation refers to the spontaneous development of oxidized flavor in milk. This flavor is a result of lipid autooxidation rather than hydrolytic rancidity and is distinct from LOF and occurs in the absence of light exposure. The off flavor is characterized by distinct metallic flavor. The exact cause of autoxidation is not well known, but mineral imbalance, cow stress, and milk fatty acid imbalance have all been hypothesized as possible contributors (Barrefors et al., 1995; Glantz et al., 2009). Autooxidized off-flavor development is often not apparent until post-processing when the milk is in the consumer supply chain (Ishler and Roberts, 2016). Anecdotal studies suggest that one farm or a few cows on one farm may be the source of autoxidation in commercial milk processing. As such, in commingled milk which is not evaluated for sensory quality until post pasteurization, this off flavor may be unnoticed until 8-10 days into the consumer shelf life when autooxidative products are at sufficient levels for sensory detection.

The process of pasteurization may reduce autoxidation through the formation of sulfhydryl compounds that can quench free radicals and/or the resulting cooked flavors may mask autoxidation flavors. As such, ultrapasteurization may provide even more mitigating or masking effects (Lee et al., 2017). While the exact cause is debated, some studies have speculated that autooxidation is likely related to vitamin E deficiency in cows during the winter and spring months. Ascribing to this theory, susceptibility of milk to autooxidation may be remedied by increasing cow access to green feed during these seasons (Barrefors et al., 1995).

Feed Flavors

Flavors related to feed or forage for dairy cows are readily apparent in milk as soon as 2-4 h post-consumption (Schiano et al., 2017). Furthermore, these flavors may be imparted into collected milk supply for as long as 24 hours post-consumption (Urbach, 1990). In general, feed flavors are quite variable and are specific to the feed quality and feed type(s), meaning that regional differences in milk flavor are common, as are differences related to farming practices. That said, dissimilarities among feeding regiments are not defined by specific flavor compounds. Volatile compound analysis of milks from pasture and total mixed ration (TMR) diets have shown that the volatile components of fluid milk are generally the same, regardless of feeding regimen and flavor differences may be attributed to differing concentrations/ratios of these components (Bendall, 2001). For conventional milk systems in which cows are fed TMR silage, milk flavor tends to be characterized by high sweet aromatic flavor and mild malty notes. In contrast, milk from pasture-based forage systems tend to exhibit grassy, cowy, barny, and mothball flavors, as well as increased salty taste (Croissant et al., 2007). Consumers are generally able to differentiate milks based on feed related flavors, but conclusions about whether these differences significantly impact acceptance or preference are unclear (Croissant et al., 2007; Harwood and Drake, 2020).

PASTEURIZATION

Pasteurization, named after French biologist Louis Pasteur, refers to the application of heat for the purposes of microbial reduction and reducing risks associated with food-borne pathogens. In industry, the application of thermal processes for microbial reduction takes on several forms, with various application techniques and time-temperature combinations being employed to achieve specific purposes. As is common throughout the food and beverage industry, application of heat under various time domains promotes significant changes to the structural and chemical constituents of the raw material to which they are applied. In the case of milk, whey, and other dairy products which are largely defined by their “clean” sensory profile, these changes may present significant and consumer-relevant alterations to sensory quality.

To meet the qualifications for a “pasteurized” product, each particle within the food system must be brought to the target temperature for a specific amount of time. Doing so facilitates adequate reduction in microbial load and enzyme deactivation, which in turn extends product shelf life from a quality perspective and, more importantly, ensures consumer safety. Under the Pasteurized Milk Ordinance (FDA, 2017), there are standards for different methods of pasteurization for milk with respect to temperature and the residence time at said temperature (Table 2).

Table 2. Time-temperature combinations for milk pasteurization

Batch (Vat) Pasteurization	
Temperature	**Time**
63 °C (145°F)	30 min
Continuous Flow (HTST and HHST) Pasteurization	
Temperature	**Time**
72°C (161°F)	15 seconds
89°C (191°F)	1.0 seconds
90°C (194°F)	0.5 seconds
94°C (201°F)	0.1 seconds
96°C (204°F)	0.05 seconds
100°C (212°F)	0.01 seconds
Ultrapasteurization	
Temperature	**Time**
138°C (280°F)	2 seconds

Adapted from the 2017 Pasteurized Milk Ordinance (FDA, 2017)

Time-temperature combinations for fluid milk pasteurization are generally defined as either batch, high temperature short time (HTST), higher heat shorter time/extended shelf life

(HHST/ESL), or ultrapasteurized/ultra-high temperature (UP/UHT). Batch or vat pasteurization (minimum 30 min. hold time at 63°C) is seldom used in modern dairy operations and is typically only used in small family farming operations. HTST (minimum 15s hold time at 72°C) and UP (minimum 2s hold time at 138°C) are the most widely utilized methods for thermal treatment of milk and dairy products in the United States. UHT treatment- an extension of UP thermal processes to include in-line aseptic fill of product is less common in US markets, but allows for ambient storage of sterile milk, which may be attractive to some consumers and manufacturers.

Differences in sensory profile of fluid milk stemming from the higher heat treatment of UP processes (compared to HTST processes) have generally been poorly accepted by American consumers of all ages (Blake et al., 1995; Chapman and Boor, 2001; Lee et al., 2017). Sensory changes associated with different thermal treatments are often noticeable in downstream processes and final commercial products, presenting consumer-relevant concerns. The flavor difference between UP milk and traditional HTST milk has been consistently described by higher cooked flavor, higher sulfur/eggy notes, higher astringency and lingering aftertase (Chapman et al., 2001; Clare et al., 2005; Valero et al., 2001; Andersson and Öste, 1995; Lee et al., 2017, Jo et al., 2018). UP milk flavors may exhibit further differences due to if the milk is processed by direct versus indirect heat treatment (Jo et al., 2018, 2019). Aseptic shelf stable milk is further characterized by more intense caramelized and cardboard flavors compared to UP milk of the same fat content (Table 3). Formation of cooked or eggy/sulfurous flavors in thermally processed milk and whey is caused by denaturation of native whey proteins- primarily β-lactoglobulin and α-lactalbumin (Jo et al., 2019). In thermal processes exceeding about 60C, whey proteins are unfolded, facilitating the liberation of volatile sulfur compounds associated with sulfur, burnt, and eggy flavors, such as hydrogen sulfide and carbon disulfide (Jo et al., 2019). The sensory differences as well as the sources of those differences between UP and aseptic milk have not been well-studied. Presumably, these differences may be due to storage time, storage temperature and or package differences (e.g. oxygen permeability) between these milks.

Table 3. Sensory differences between ultrapasteurized (UP) and aseptic fluid milks

Attribute	Aseptic 1	Aseptic 2	UP
Aroma Intensity	2.3a	2.4a	2.9a
Aroma Character	Caramel	Caramel, stale	Cooked/sulfur
Sweet Aromatic	1.7a	ND	1.1b
Cooked/Milky	4.4a	4.0b	4.4a
Sulfur	ND	ND	2.4
Caramelized	1.5b	3.2a	ND
Cardboard	1.4b	2.7a	ND
Sweet Taste	2.8a	2.9a	2.3b
Salty Taste	2.2b	2.9a	2.2b
Astringency	3.0b	3.3a	2.6c
Viscosity	2.3a	2.3a	2.2b
Comments	caramelized	Caramelized, stale	Cooked sulfur

*Different letters in rows indicate a significant difference ($p<0.05$). ND – not detected. Attributes were scored using a 0 to 15 point scale consistent with the Spectrum descriptive analysis method. Most fluid milk flavors fall between 0 and 4 on this scale (Lee et al., 2017).

The prominence of cooked and sulfur flavors resulting from greater thermal treatment may additionally be affected by intrinsic raw product qualities such as milkfat. Observations by Valero et al. (2001) showed that these off-flavors were significantly higher in skim milk than milk with 2% milkfat. These findings were corroborated by Jo et al. (2018), who noted a significant interaction between milkfat and thermal process on the formation of cooked flavors in fluid milks. Similarly, Oupadissakoon et al. (2009) demonstrated that common dairy attributes such as dairy flavor, dairy fat, and dairy sweetness were negatively correlated with chalky texture and processed or cooked flavors among different heat treatments.

Besides flavor, a significant point of difference between HTST and UP processed products is viscosity, which is measured as the perceived thickness or mouthfeel of the product.

In thermal processes exceeding about 60-70°C, native β-lactoglobulin denaturation promotes formation of complexes with other denatured β-lactoglobulins or casein micelles, thus increasing particle size and viscosity (Li et al., 2018). As such, whey protein concentration during thermal processing can play a significant role in viscosity change, with higher protein concentrations being associated with greater increases in viscosity (Singh et al., 2019). For fluid milk, these changes are rather mild, and may even enhance consumer perceptions. In a study by Chapman et al. (2001) with trained panelists, viscosity ratings for nonfat UP milks were approximately the same as for 1% HTST milks. Similarly, these findings were corroborated in consumer milk evaluations by Lee et al. (2017), with adult consumers reporting UP-treated skim milks (both direct and indirect heating methods) were noticeably thicker than HTST skim milk. However, the viscosity/mouthfeel differences between treatments were not as readily noticed by children in the same study. Overall, these findings conceptually bode well for UP milks because consumers, in general, prefer the viscosity of 2% fat milks to those with lower fat contents (Pangborn, 1985). UP processes may also contribute undesirable astringent mouthfeel characteristics which are similarly associated with protein denaturation in fluid milk. Higher astringency in UP milk treatments has been noted by trained panel immediately following thermal processing, with the effects sustaining through several weeks of storage (Li et al., 2018).

While ultrapasteurized milk, cream, and whey products are common in European, African, and Asian countries, they are comparatively less ubiquitous in the United States and have been generally viewed as less preferred (compared to HTST-treated products) by American consumers. Past explanations for the preference of HTST milk over UP milk include familiarity (Chapman and Boor, 2001) and the presence of sulfurous off-flavors (Valero et al., 2001). Evaluation of Chinese and Australian milk consumers demonstrated that as expected, Chinese consumers preferred shelf stable milks while Australian consumers preferred HTST milks, but Liem et al. (2016) further noted that flavor differences in HTST and UP milks may implicitly influence perceptions related to product quality and safety, solidifying differences in ultimate acceptance. While these preferences have been shown to exist, there is evidence that flavors associated with UP milks are not unacceptable altogether. In tasting experiments with American children, Chapman and Boor (2001) found that traditional HTST milk was preferred over UP and UHT milk, although the average degree of liking on a 7-point hedonic scale of UP milk was found to be only slightly below "good" (4.7) indicating baseline acceptability. Similarly, Lee et al. (2017) also noted that children preferred HTST milk over UP milk, but liking scores (7-point hedonic scale) of UP milks reported by children were relatively equal across fat contents and heat transfer techniques (scores between 4.4 and 4.7 for skim and 2% milks from direct or indirect UP treatments). Consumer acceptance testing on HTST milks subjected to a range of time/temperature treatments followed by cluster analysis by Gandy et al. (2008), demonstrated that some people liked milk with cooked notes, while other people did not like the cooked notes, suggesting that generalized commentary on UP milk acceptability may be somewhat misleading. It is entirely possible that the sensory divide between UP milks and traditional HTST milks in the US market is being lessened as a result of increased organic milk consumption. Commercial organic milks are often ultrapasteurized. As such, it is possible that negative consumer sentiments for UP products are being assuaged as a function of increased familiarity.

Direct vs. Indirect Heat Exchangers

The application of either direct or indirect heating systems can dictate the presence and intensity of specific sensory features associated with ultrapasteurization, warranting additional consideration by dairy processors. Indirect heating systems heat product through a partition separating the raw food material stream from the heating medium (commonly steam or water). Indirect systems may take the form of tubular or plate heat exchangers in continuous flow processing operations. On the other hand, direct heating involves the direct contact of raw product with the heating medium, which is typically culinary steam in the case of dairy processing. Within a direct heating system, the combination of raw product and culinary steam may be achieved through direct steam injection (DSI) or infusion of raw material into steam, with the former being most common. By nature, direct heating introduces water into the food system, diluting the product; as such, direct heating systems generally necessitate subsequent processing steps to remove added water from the system and cool the product. The most common system for these purposes is flash vacuum cooling, whereby heated product with added water is introduced to a vacuum chamber. Within the vacuum chamber, applied pressure is quickly decreased, vaporizing the water in the product. A vacuum is concurrently pulled within the chamber, removing water vapor and quickly decreasing product temperature (Lee et al., 2017).

Relevant sensory differences stemming from the use of either indirect or direct heating systems are numerous. Localized heating at the indirect heating interface of tubular and plate heat exchangers is associated with increased amounts of “fouling” due to protein denaturation and burn-on (Datta et al., 2002). This increased rate of protein denaturation in indirect systems leads to comparatively greater increases in viscosity than direct heating for fluid milk and whey

(Lee et al., 2017; Kelleher et al., 2018). Direct heating of dairy products is generally viewed as advantageous to indirect in terms of heat load, energy efficiency, and sensory profile (Lee et al., 2017; Rattray et al., 1997; Figure 2). Denatured proteins have flavor binding qualities. Since direct heating systems imbue lesser thermal load than indirect systems, the degree of protein denaturation is significantly decreased. This results in increased intensity of volatile sulfur flavors formed from high heat thermal processes (Jo et al., 2018). As previously mentioned, such flavors are generally disliked by consumers, requiring dairy products processed using direct UP or UHT to be stored until such flavors subside.

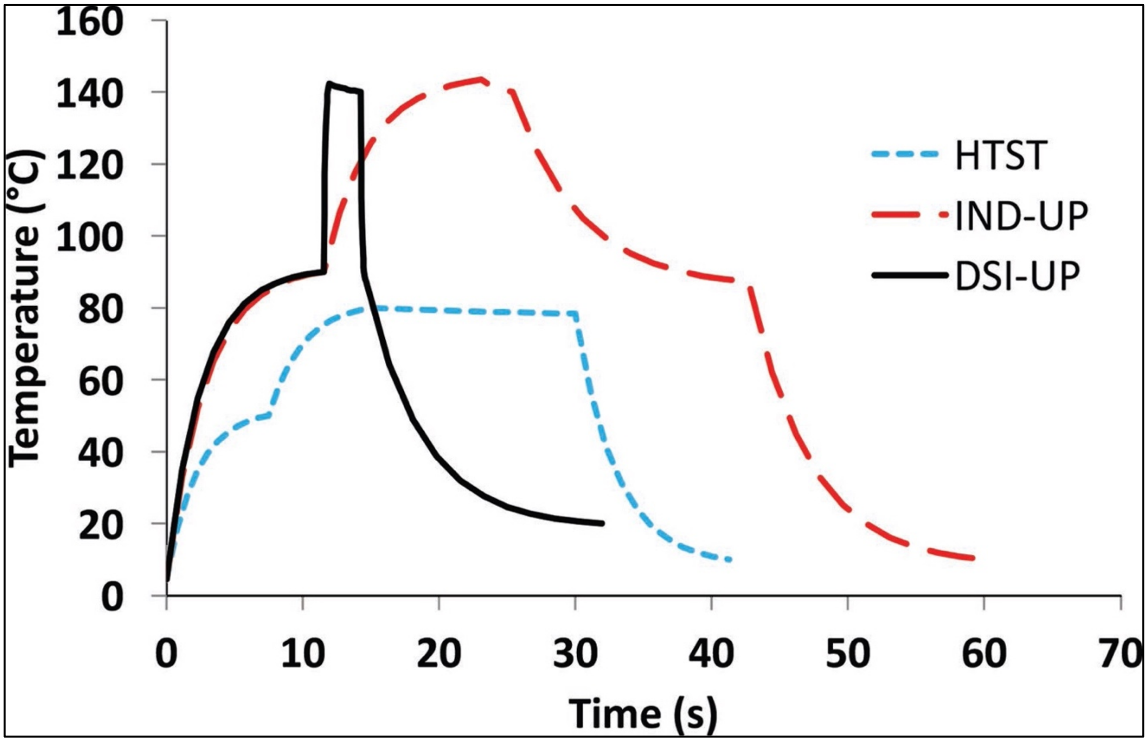

Figure 2. Thermal profiles of HTST pasteurized milk and ultrapasteurized (UP) milks processed using indirect (IND) or direct steam injection (DSI) heating methods
Taken from Lee et al., 2017

Storage of Pasteurized Milk

Different time-temperature combinations for thermal processing result in different shelf lives for milk and dairy products- primarily a function of the increased inactivation of spoilage bacteria at elevated temperatures. In refrigerated storage condition, HTST milk has a shelf life around 21 days under refrigeration, UP milk up to 70 days under refrigeration and UHT milk has a shelf life of up to nine months at ambient conditions (Tay and Chua, 2015). Microbial growth and metabolism shorten the shelf life of milk by producing undesirable changes in aroma and taste attributes that influence consumer acceptability of the products. Additionally, non-spoilage related changes to product taste and quality should be considered. Lipid oxidation occurs in fluid heat treated milk during storage. If microbial processes do not predominate, lipid oxidation results in decreased fresh sweet aromatic and cooked flavors and increases in cardboard and stale flavors. Rate of lipid oxidation is mediated largely by processing methods employed, packaging, and storage temperature. This is especially relevant for products such as aseptic milk. Aseptic is subjected to thermal treatment the same as, or similar to, UP processes; however, aseptic milk is additionally filled under sterile conditions into brick or prisma packages allowing for storage under ambient conditions. This packaging is less oxygen permeable than the gabletop paperboard cartons used for UP processed milk, thus mitigating stale flavor development (Mehta and Bassette, 1978).

Some studies have shown beneficial effects of storage related to high-heat thermal processes for fluid milks. For example, sulfurous, burnt, and eggy off-flavors that lead to low consumer acceptance (Chapman and Boor, 2001; Gandy et al., 2008) decreased with 14 days of refrigerated storage time (Jo et al., 2018). Further refrigerated storage of UP skim and 2% fat milk results in further decreases in sulfur/eggy flavors and cooked flavors, but HTST milk remains distinct in flavor from UP milk (Figure 3) and was still preferred by adult US consumers over UP milk, even following 45 days of refrigerated storage (Figures 3 and 4). Findings by Harwood et al. (2020) recently demonstrated that UP treatments were less susceptible than HTST milks to light oxidation flavor development that may occur during commercial refrigerated storage. Considering these qualities, UP and other high-heat processed milks may present unique advantages over their HTST counterparts during commercial (in-grocery) storage/display in terms of quality preservation.

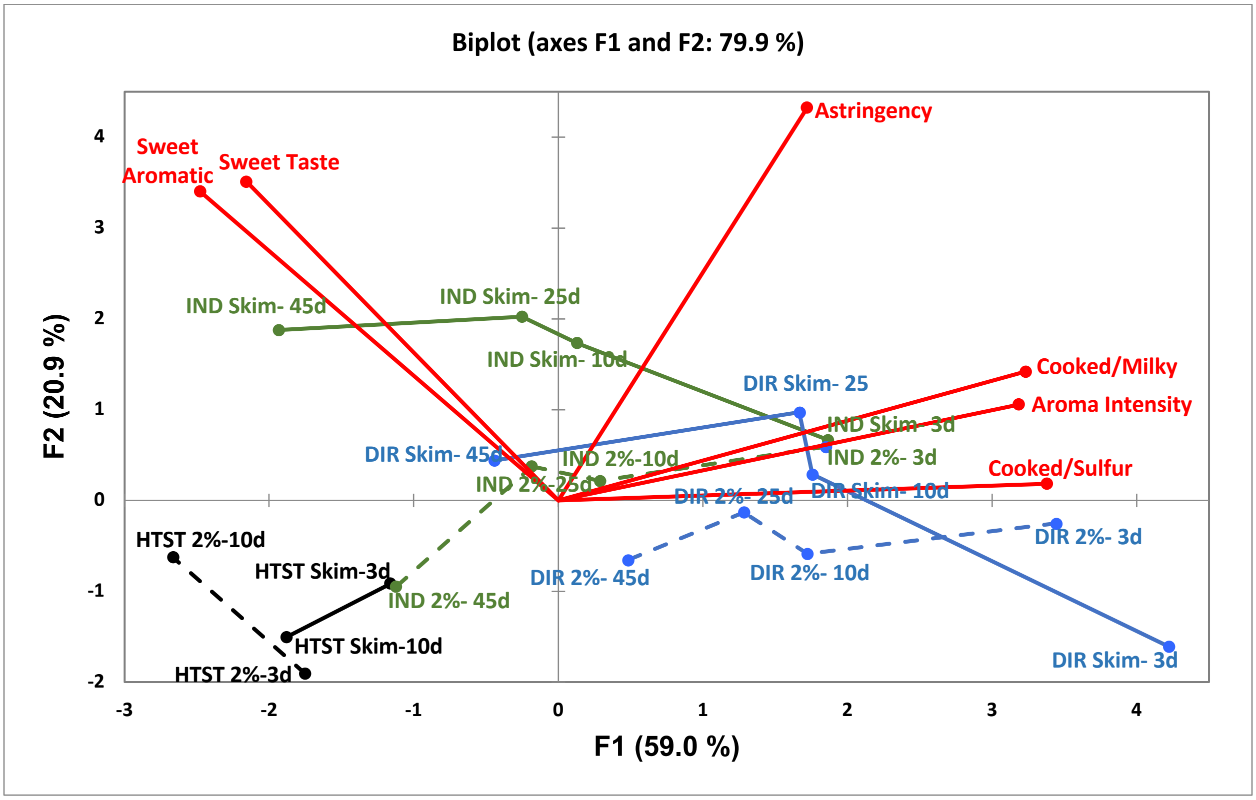

Figure 3. Dissipation of cooked and sulfur flavors during refrigerated storage for direct steam injection (DSI) and indirect (IND) ultrapasteurized (UP) skim and 2% milks

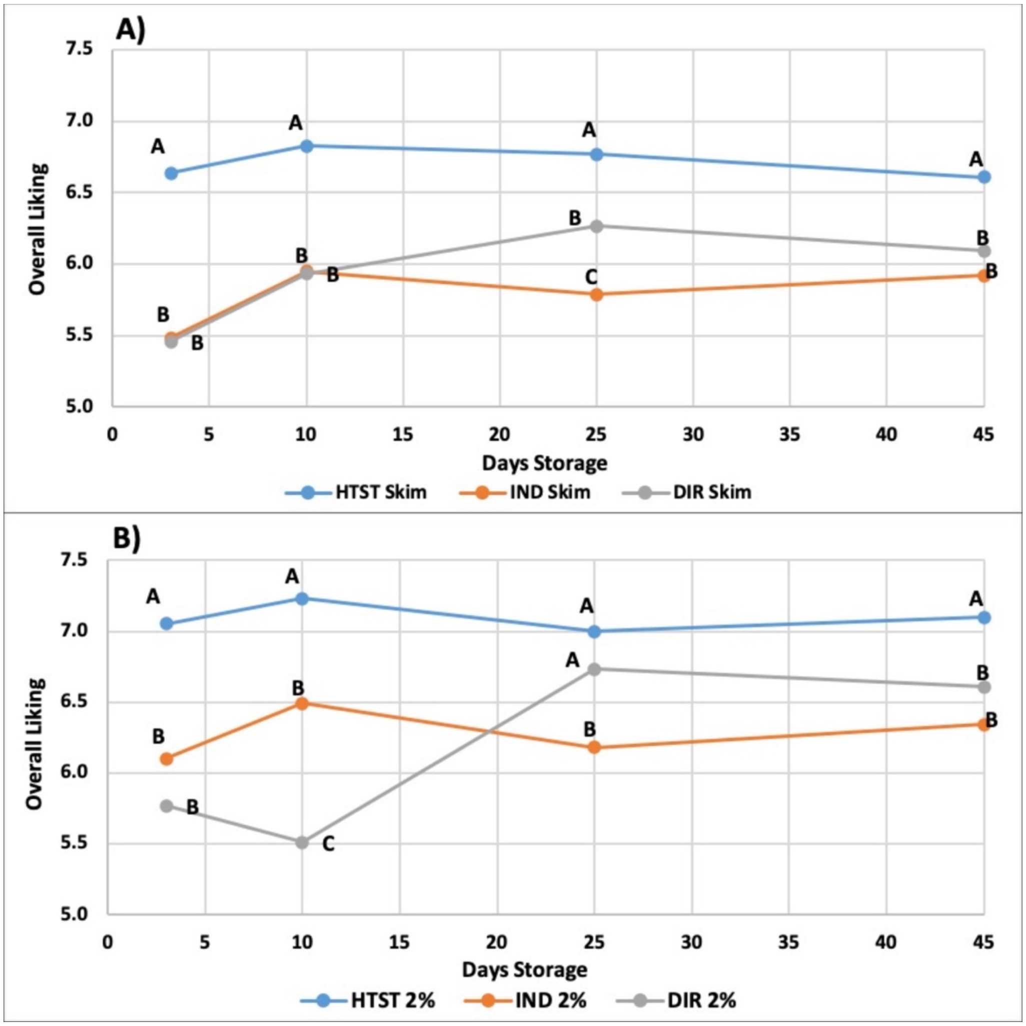

Figure 4. Consumer acceptance scores of HTST, DSI-UP, and IND-UP skim and 2% milks during refrigerated storage. Overall liking was scored on a 1 to 9 point intensity scale where 1 = dislike extremely and 9 = like extremely. At each timepoint, each milk was evaluated by 100 fluid milk consumers of that particular fat content (eg skim consumers vs 2%/whole milk consumers). After day 10, subsequent consumer evaluations at each timepoint were compared with 10 day old HTST milk

While some storage-related benefits for ESL/UP/UHT products may exist, off-flavor development and quality deterioration during extended storage should be chiefly considered. Blake et al. (1995) found that extended shelf life whole milks processed using direct steam injection showed a significant decrease in cooked flavor after 15 days storage time at 7°C, but also exhibited increases in stale off-flavors. In addition to stale/oxidized flavor development, other sensorial changes may be noted during milk storage. For example, elevated storage temperature of shelf stable milk is associated with darkening of product color and accelerated formation of caramelized and Maillard off-flavors that may only form slowly or not at all within a typical shelf life at ambient or refrigerated conditions (Koffi et al., 2015; Sunds et al., 2018). Additionally, UP/UHT thermal processes alone may be insufficient in the inactivation of plasmin- a native milk protease- resulting in sedimentation, separation, or gelation for shelf stable milk and whey products (Chavan et al., 2011). This is especially the case for direct method UHT milk, where gelation and bitter taste development have been cited concerns (Nursten, 1997). This is likely a function of decreased inactivation of proteases stemming from lower thermal load of direct UHT heating, as compared to indirect heating.

Alternative Pasteurization Techniques

Retort

In-package thermal processing for the purposes of product sterilization may be considered for milk and dairy products. Advantages of retort treatment are similar to those in UHT systems- increased shelf life and ability to store products at ambient temperatures. However, negative sensory effects related to retort processes have also been reported. Dairy products that undergo retort are often subject to considerable heat loads (up to 120°C for up to 40 min.); as such, pronounced cooked flavors and Maillard browning are common sensory features of retorted products (Boor and Murphy, 2002). Vitamin losses and decreased freshness have also been noted as common disadvantages related to retort of milk-based products (Rapaille and Vanhemelrijck, 1998). Additionally, the prolonged shelf life associated with retort may present quality defects. Evaluations by Cano-Ruiz and Richter (1998) showed that gelation and settling were relevant sensory problems in retorted dairy formulations during extended ambient storage.

Microwave Heating

Utilization of microwaves for the purpose of heating milk and dairy products in continuous systems has been proposed as a viable pasteurization option for processors (Valero et al., 2000). From a sensory perspective, microwave sterilization of white milk was shown by Clare et al. (2005) to maintain lighter color and produce milder cooked/caramelized flavor compared to indirect UHT treatment. Furthermore, triangle tests reported by Jaynes (1975) comparing microwave pasteurized milk (72°C for 15s) to vat pasteurized milk (63°C for 30 min.) showed no significant sensory differences were present between the two products. While the sensory and nutritional advantages of microwave processing make it an attractive alternative to traditional pasteurization systems in the dairy industry, issues related to non-uniform temperature distribution in continuous systems, costliness, and energy efficiency often prohibit the use of such technologies (Melini et al., 2017).

High Pressure Processing

High pressure processing (HPP) has been proposed as an alternative to thermal treatments for the inactivation of enzymes and bacteria in fluid milk and is allowable under the PMO assuming pathogen reduction is akin to pasteurization (FDA, 2017). As its name suggests, HPP employs heightened pressures, between 100 and 1000 MPa, to disrupt secondary and tertiary protein structures within a given food system (Voigt et al., 2015). Compared to thermal processes, HPP is superior in retention of flavor, texture, and nutritional profile in products such as cheese or yogurt (Rastogi et al., 2007). Furthermore, HPP does not affect smaller components within milk and dairy products, such as amino acids or added flavor compounds (Balci and Wilbey, 1999). For certain cheese types, circumventing thermal processes is of particular interest, as milk pasteurization flavors and the process of pasteurization can impact cheese sensory attributes and cheese ripening; however, investigation of the differences in consumer acceptance between cheeses from thermally processed milk and HP processed milk have been inconclusive (Drake et al., 1997; Buffa et al., 2001).

Sub-Processes

In a typical dairy operation, pasteurization is applied in conjunction with several other sub-processes that affect milk sensory quality. Among the most common processes that appear in-line with continuous pasteurization systems are deaeration and homogenization. While these methods are relatively mild in their sensory contributions to milk and dairy product, each confer qualities that may influence downstream processing effects.

Deaeration/Degassing

Milk naturally has some measure of dissolved gases from the time it is collected. The volume gas content within collected milk is directly related to the oxygen in the cow's bloodstream and may be between 4.5% and 6% at the time of milking (Anonymous, 2003). Following fluid milk collection on the farm, raw milk is typically subjected to several handling and transportation steps before finally reaching a site for processing. During this time, fluid milk is agitated and exposed to atmospheric oxygen, allowing for the infusion of dissolved gases. High concentrations of dispersed air in fluid milk can present a number of complications related to product quality if left unaddressed. Dissolved oxygen in fluid milk and milk products has been associated with higher redox potential post-processing (Giroux et al., 2008), vitamin loss (Oamen et al., 1989), and reduction of free sulfhydryl groups (Perkins et al., 2007). The process of deaeration may also reduce feed-related flavors in raw milk that may contribute to off flavors (Bassette et al., 1986). As such, reduction of dissolved oxygen within the fluid milk processing line is often done in order to ensure downstream product quality. In practice, oxygen removal in a continuous pasteurization system is introduced following preheating. In this process, preheated product is introduced to and expansion chamber at a decreased pressure. The combination of heat and a sudden pressure drop lowers vapor pressure in the system, expelling water and dissolved gases within the system. Next, a built-in condenser captures the water vapor, allowing for the water to be condensed and returned to the milk, while a vacuum system removes the non-condensable gases and volatiles in the system. While the general purpose of degassing in fluid milk processing is oriented at gas removal, milk used in certain products, such as yogurt, may forego condensation steps, allowing for mild concentration (Anonymous, 2003). Similarly, degassing approaches that waive condenser use mirror the mechanisms used in flash vacuum cooling of UHT milk and whey, explaining the relatively low dissolved oxygen values inherent to such products (Harwood and Drake, 2020).

From a processing standpoint, removal of dissolved oxygen is chiefly oriented towards maintaining product quality during storage; however, the removal of volatile compounds that coincides with vacuum systems in dairy processing may alter product sensory qualities in other ways as well. Carter (2020) evaluated the volatile compounds removed by vacuum cooling in DSI-UP fluid milk and found that many aroma-active compounds were removed by this process.

Homogenization

Homogenization refers to the application of high pressure and shear for the purpose of decreasing fat globule size, thus ensuring uniform dispersion of milkfat and preventing milkfat coalescence. While some exceptions exist in local or organic commercial milks, the vast majority of market milks are homogenized (Alvarez, 2009). Milkfat dispersion resulting from homogenization has several effects on milk quality and susceptibility to off-flavor development. In particular, homogenized milks have been reported to be less susceptible to spontaneous autooxidation (Walstra and Jenness, 1984), and whole milk powders made from homogenized milks are less susceptible to cardboard and painty off-flavor development (Park and Drake, 2017). Additionally, homogenization imparts direct alterations to fluid milk mouthfeel and appearance, resulting from increased milkfat dispersion and the light-scattering effect thereof (Pritchard and Kailasapathy, 2011). These effects are similarly carried through in downstream processes for dairy products such as cheese (Madadlou et al., 2007) and coffee creamers (Chung et al., 2017).

WHEY PROCESSING

Whey protein refers to the native soluble serum proteins in fluid milk, including α-lactalbumin, β-lactoglobulin, serum albumin, and immunoglobulins (Anonymous, 2003). Current nomenclature refers to these soluble proteins as whey proteins when they are separated from cheese whey. In contrast, when these proteins are present in fluid milk or separated directly from fluid milk without a cheese-make or acid coagulation process, they are also referred to as milk whey proteins. Separation of whey proteins from cheese whey and more currently directly from milk is used to produce commercial whey protein concentrates (WPCs; 34-80% protein) or isolates (WPI; >90% protein) (Foegeding and Luck, 2011). Quality and sensory changes related to filtration and concentration of whey protein fractions will be discussed in Section 5 of this chapter. However, variables within the traditional cheese make method for whey protein production may also affect product attributes and should be considered.

Rennet and Starter Cultures

Liquid whey has distinctive flavors which vary depending on the starter cultures and cheese make-procedure. These flavors from the initial whey formation have a direct influence on the flavor of whey protein or other whey ingredients manufactured from these sources (Smith et al., 2016a, 2016b, 2016c). In general, whey streams may be described as either “sweet whey” or “acid whey”. Sweet whey refers to whey produced as a byproduct of rennet-coagulated cheeses, whereas acid whey refers to whey produced via acid-coagulated cheeses (Tunick, 2008). Utilization of rennet alone for cheesemaking results in a product that is characteristically bland, sweet, milky, and free from off-flavors (Smith et al., 2016a); however, the use of different starter cultures in addition to rennet has been shown to impart cardboard and other flavors (Gallardo-Escamilla et al., 2005; Campbell et al., 2011a). The impact of different starter cultures on the flavor of sweet whey is variable and impacted by mesophilic versus thermophilic starter cultures as well as different strains within a type of starter culture (Liaw et al., 2011; Campbell et al., 2011a, 2011b). Acid whey, on the other hand, is defined by sour, salty, and bitter tastes, as well as high astringency and sour aromatic flavors (McGugan et al., 1979; Gallardo-Escamilla et al., 2005; Smith et al., 2016a).

Whey Bleaching

The use of annatto or other natural colorants in the production of yellow or orange cheese types such as Cheddar and Colby, necessitates color remediation of liquid whey since consumers expect these cheeses to have an orange color. Among the most common bleaching agents for these purposes are chemical agents: benzoyl peroxide and hydrogen peroxide, and enzymatic bleaching (Carter and Drake, 2018) although activated charcoal has also shown promise (Zhang et al., 2015). As is intended, bleaching agents are generally successful in creating more acceptable color for liquid whey protein (Kang et al., 2010; Zhang et al., 2015); however, bleaching is an oxidative process and produces lipid oxidation and protein degradation products which constitute a primary source of off flavors in bleached whey and ingredients made from bleached whey (Jervis et al., 2015; Carter and Drake, 2018).

Fluid Storage of Whey

Following whey manufacture and subsequent concentration steps (discussed below), liquid product is typically held for up to 48h under refrigerated storage before being spray dried or otherwise processed as an ingredient. During this transition time, significant alterations to whey flavor may be imparted to the liquid product, and these flavors carry through to downstream products. In the case of spray dried WPC and WPI, evolution of flavor is largely dependent on storage and handling pre-spray drying. Development of off flavors during liquid storage is largely due to lipid oxidation. Observations by Whitson et al. (2011) showed that increased liquid storage time before spray drying was associated with greater intensities of astringency and overall aroma as well as cardboard and serumy flavors in mozzarella whey protein concentrate. Similarly, increased aroma intensity, cardboard, and fatty notes were found for Cheddar WPI when whey protein retentate was stored for 48h, compared to product from fluid retentate that was not stored. It should be noted that while off flavor development during storage of liquid whey is a constant for all whey types, sensory profiles of different starter cultures and strains are quite unique and should be independently considered in terms of susceptibility to lipid oxidation (Campbell et al., 2011b; Smith et al., 2016a). To mitigate lipid oxidation effects in liquid whey, defatting processes and addition of hydrolysates or other antioxidants have shown modest success (Liaw et al., 2010). Furthermore, manufacturers may consider timing of pretreatment steps, as concentration early in the storage process and bleaching later in storage beneficially impedes lipid oxidation and off flavor development of liquid whey (Park et al., 2016a).

CONCENTRATION AND FILTRATION OF MILK AND WHEY

Removal of water from liquid milk and whey systems can serve a variety of purposes. Water adds to bulk weight; thus, its removal may play a practical and economic role in ingredient transportation. Processes that facilitate water removal are often oriented towards improving stability, or to reducing susceptibility to bacterial spoilage- a phenomena often linked to moisture content. Additionally, concentration of a given material stream may be a necessary processing step to satisfy requirements for product identity- as is the case in WPC and WPI manufacture. Regardless of the specific process employed, concentration and filtration significantly alter product quality, functionality, and identity, warranting greater understanding from both processors and consumers alike.

Evaporation

Evaporative dehydration as a means for product concentration is commonly applied to milk and milk concentrates but may be employed as a means for concentration (40-60% solids) prior to spray drying for liquid whey streams as well (Pearce, 1992). Direct sensory influences to dairy products following falling film evaporation under vacuum include increased viscosity and increased sweet and salty tastes as a result of heightened solid content. Cooked flavors related to thermal load in the evaporative process may also be exacerbated by changes in concentration. This is especially true for products containing lactose, as increased concentrations of protein and lactose encourage Maillard browning. As a result, condensed milk products generally exhibit significant darkened color, as well as cooked, cardboard, and caramelized flavors following evaporation and subsequent thermal processing (Drake et al., 2003; Hwang et al., 2006; Park and Drake, 2016). Furthermore, continuation of Maillard browning processes during storage may enhance caramelized flavor and darken product color (Hwang et al., 2006; Sharma et al., 2015).

The vacuum processes used to facilitate vapor removal in the evaporator system have been shown to remove volatile compounds in milk, significantly reducing sweet aromatic flavor impact (Park and Drake, 2016). Park and Drake (2016) demonstrated that alternative methods for production of condensed milk and whey, such as partial water removal via reverse osmosis, should be utilized when possible to preserve product sensory quality.

Membrane Filtration

Membrane technology in milk and whey processing refers to the use of semi-permeable membranes for the purposes of selectively filtering, fractionating, or concentrating targeted components (Ho and Sirkar, 1992). These processes typically include, ultrafiltration, microfiltration, nanofiltration, and reverse osmosis, and may be aided by diafiltration in order to further refine the product stream. Filtration methods are differentiated by pore size and may be selectively employed to meet process needs. Direct contributions to flavor are rare in membrane filtration systems. Soapy and detergent off flavors have been attributed to fluid milk and whey ingredients due to inadequate rinsing of membranes post-cleaning. It is also possible and logical that dirty or fouled membranes may contribute off flavors. In contrast, water removal and or solids concentration by membrane processing compared to other methods may result in lower lipid oxidation and cooked/Maillard reactions which results in improved flavor. Removal of water by reverse osmosis prior to spray drying has been shown to produce a higher sensory quality milk powder compared to milk powder produced by evaporation prior to spray drying (Park and Drake, 2016). A higher solids concentration prior to spray drying improves the flavor of whey and milk products compared to lower solids at spray drying (Park et al., 2014a).

Although direct alterations to flavor are scarce, filtration and concentration, by design alter the component ratios of milk and whey, resulting in products with distinct sensory characteristics simply by concentration or removal of specific milk and whey components. Ultrafiltration, for example, has been suggested as a viable means for lactose and mineral removal and protein concentration in fluid milks. However, lactose-free milks resulting from lactose removal are characteristically less sweet and lower in salty taste than those produced via enzymatic hydrolysis of lactose (Rizzo et al., 2020). Sensory differences associated with enhanced protein concentration have also been noted in whey and milk protein streams. As protein concentration increases and lactose/mineral content decreases in milk and whey protein concentrates and isolates, cooked/milky and sweet aromatic flavors decrease while protein and process-specific flavors animal, cardboard, brothy, and tortilla flavors increase (Drake et al., 2009; Smith et al., 2016d). Furthermore, alteration of whey/casein ratios in fluid milk beverages, MPC, and micellar casein concentrates can impact functional properties but also affect visual, flavor and mouthfeel properties (Renhe and Corredig, 2018; Cheng et al., 2019a, 2019b).

Spray Drying

The manufacture of milk and whey powders via spray drying presents practical advantages related to product transportation and shelf life, however, sensory changes resulting from drying processes have been widely reported. In general, spray dried powders are characterized by higher aroma impacts and greater oxidized off-flavors compared to liquid ingredients of the same type (Carter et al., 2018). Further sensory changes resulting from spray drying techniques are largely determined by the combinatorial effects of feed solid concentration and inlet temperature. Experiments by Park et al. (2016b) demonstrated that spray drying of condensed skim milk and MPC70 at higher inlet temperatures (260C) resulted in powders with lessened oxidized/cardboard sensory character compared to lower inlet temperature (160C). Conversely, higher inlet temperatures were associated with higher sweet aromatic flavors and overall aroma impact, an indication of greater Maillard browning. In terms of feed concentration, low feed solids (10% solids) for spray dried Cheddar WPC80 were shown by Park et al. (2014a) to result in powder with decreased particle size and increased surface fat. These structural features of low feed concentrations have been linked to greater oxidative potential and increased formation of cardboard and cabbage off-flavors compared to powders with increased particle sizes and decreased surface fat resulting from higher feed concentrations (Park and Drake, 2014; Park et al., 2014a).

For milk and whey products, sensory effects resulting from spray drying may additionally be affected by preceding processing steps. For example, color changes and formation of sweet aromatic and caramel flavors resulting from Maillard browning may be mitigated by lactose removal following ultrafiltration. As such, milk protein isolates, whey protein isolates, caseins, and caseinates are of comparatively less risk than milk powders or whey concentrates to Maillard-related sensory changes during spray drying and powder storage (Higgs and Boland, 2014). Additionally, Park et al. (2014b) showed that acidification of liquid whey protein (WPC80) pre-spray drying modulated off-flavor development in the spray dried protein powder. While cardboard flavor and lipid oxidation products were widely reduced by acidification, it should be noted that rehydrated acidified WPC80 was found to be characteristically soapy. In summation, the harsh heating and dehydrative effects of spray drying should be considered pre-processing to ensure adequate sensory profiles of the resulting powders are maintained.

Powder Storage

Milk and whey powders are generally characterized by high stability over shelf life. This is especially the case in situations where ambient storage conditions, free from temperature abuse and high moisture/humidity, can be maintained. While dairy powders are largely more stable during storage than their liquid counterparts, monitoring off-flavor development is important because these flavors are typically carried through in various ingredient applications and may be readily noticed by consumers, thus decreasing acceptance (Caudle et al., 2005). Susceptibility of powders to flavor and quality changes over time is largely product dependent. Powders with high fat content, such as whole milk powder (WMP) have a comparatively shorter shelf life than products with lesser fat, such as SMP (Drake et al., 2006; Carunchia-Whetstine and Drake, 2007). This is primarily a function of greater susceptibility to lipid oxidation during storage. Flavor degradation of WMP following extended ambient storage is characterized by decreases in cooked/sulfur and fresh sweet aromatic notes, as well as increased off-flavors including increased astringency and grassy, fatty, and painty flavors (Carunchia-Whetstine and Drake, 2007; Lloyd et al., 2009a). Lower storage temperatures and additional packaging steps, such as nitrogen flushing have been proposed to lessen these sensory changes in WMP during storage (Lloyd et al., 2009b). SMP exhibits several similar features but is typically less inclined to form painty off flavors, and more inclined to develop cardboard, fatty/fryer oil, grape/tortilla, or, in the case of international powders, animal-like off notes (Drake et al., 2003; Carunchia-Whetstine and Drake, 2007). These flavors may appear after as little as 2 months of ambient storage, and generally increase linearly over time.

Powder concentrates and isolates from whey or milk proteins are similarly characterized by the development of off-flavors resulting from lipid oxidation and non-enzymatic browning during storage. Storage experiments by Smith et al. (2016b) have shown that MPC may develop characteristic burnt sugar, cabbage, animal, and tortilla flavors after 6 months storage, additionally, refrigerated storage (3C) may significantly delay off-flavor development whereas elevated temperature storage (40C) resulted in noticeable flavor development after only one month. In addition to flavor degradation, concurrent lapses in product quality associated with decreased solubility have been noted in stored MPC, but WPC has been reported as generally stable to such physical property effects (Anema et al., 2006; Le et al., 2011; Smith et al., 2016b).

Regardless of powder type, additional processing measures and product features should also be considered, as they influence off-flavor development rate. For example, secondary processing measures such as instantizing or agglomeration have been widely shown to decrease product shelf life. Wright et al. (2009) showed that steam and lecithin agglomerated WPC80 and WPI powders more readily formed cardboard, cucumber, and fatty flavors associated with lipid oxidation than non-agglomerated powders. Furthermore, these changes influenced consumer-relevant detractions in acceptance when applied to flavored protein beverage formulations, with agglomerated powders scoring significantly lower than non-agglomerated powders at storage times greater than 12 months. Additionally, increased moisture content and water activity in dairy powders has been shown to increase reaction rates of oxidative and non-enzymatic browning processes, thus accelerating quality and off-flavor deterioration during storage. Hygroscopic qualities of spray dried milk and whey powders make them particularly susceptible to increased moisture content during storage and several studies have reported significant increases in moisture and water activity following extended powder storage (Sithole et al., 2005; Sithole et al., 2006; Fitzpatrick et al., 2007a). In addition to typical oxidation and browning off-flavor development, loss of product functionality related to caking, decreased flowability, and decreased dispersibility is of primary concern for powders, as these features may be negatively perceived by consumers (Fitzpatrick et al., 2007b). In turn, manufacturers should give the utmost attention to packaging materials used for powders as well as storage conditions including temperature and humidity.

CONCLUSION

The various processes applied to milk and whey are a necessary means for manufacturing products that meet diverse needs, while also adhering to the utmost standards of quality and safety. However, the delicate natural sensory profile of dairy products warrants a thorough understanding of these processes and their influences on flavor and texture perception. Deeper understanding of the connections between these processes and their sensory effects serves to ensure product innovations continue toward improved product quality, and also serves as a resource for troubleshooting existing methods. Furthermore, reinforcing processes that deliver heightened consumer sensory acceptance will help to ensure success within the dairy industry for years to come.

ACKNOWLEDGEMENTS

Funding provided in part by the National Dairy Council (Rosemont, IL).

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CHAPTER 2: Validation of Fluid Milk Consumer Segments Using Qualitative Multivariate Analysis

Validation of Fluid Milk Consumer Segments Using Qualitative Multivariate Analysis

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The content of this chapter was published in:

Journal of Dairy Science (in press)

ABSTRACT

Consumption of fluid milk in the United States has declined in recent years. To increase appeal and meet ever-changing consumer demands, several product features have been introduced to the fluid milk market. As such, it is imperative to assess consumer sentiments from both quantitative and qualitative perspectives to better understand the impact of various product offerings. The objectives of this study were to identify fluid milk consumer segments that were characterized by preference for specific product features and to verify those sentiments using Qualitative Multivariate Analysis (QMA). An adaptive choice-based conjoint (ACBC) survey (n= 719) was designed to explore consumer desires regarding the fat content, package type, shelf life, label claims, and prices of commercial milks. Part-worth utilities from the conjoint task were subsequently clustered, revealing four unique consumer segments. Representative consumers from each cluster (n=18 total) were selected to participate in a 4-week QMA study, consisting of a home usage test followed by focus groups and projective mapping (PM). Nine commercial milks representing various pasteurization methods, label claims, and packaging types were used within the study. When analyzed by segment, significant ($p<0.05$) differences in conjoint utilities for specific product features were identified. Price was, overall, the largest differentiator of segments, followed by fat content, shelf life, and milk designation. Several of the segmental differences were confirmed in the QMA study, with sentiments regarding price and milk type (conventional, organic, local, etc.) consistent between conjoint survey and QMA for each consumer group. Within the PM exercise, intrinsic sensory properties, price, and quality perception were the primary differentiators for fluid milks. Comments from QMA journal entries and focus groups revealed that consumer segments with preferences for non-conventional milk types were primarily motivated by the belief that organic, local, or pasture-raised milks were

superior in sensory quality and congruent with personal values. Overall, QMA was an effective means for verifying conjoint-derived consumer groups and provided a contextual support for conjoint insights. Joint conclusions drawn from the components of this study may serve to guide marketing campaigns and new-product development for fluid milk processors.

INTERPRETIVE SUMMARY

Consumer purchase decisions are driven by both implicit and explicit product preferences. Previous consumer tests on fluid milk have generally focused on either implicit prediction methods for consumer acceptance, or solely addressed consumer sensory acceptance. However, the two methods have rarely been used in conjunction to understand consumer attitudes and preferences. This study used qualitative sensory techniques to validate survey-derived consumer segmentation in the fluid milk category.

Keywords: Milk, Focus Groups, QMA, Conjoint

INTRODUCTION

In order to counter decreased interest in fluid milk, it is imperative to understand what features consumers value in fluid milk offerings. However, there has been limited research on consumer motivations and beliefs behind commercial milk preference and purchase. Instead, the bulk of existing research has been oriented at understanding the effect of intrinsic quality features on consumer approval of fluid milk, such as feed source (Croissant et al., 2007), fat content and appearance (McCarthy et al., 2017a), and off-flavor presence (Hough et al., 2004). In order to assess these qualities, fluid milk and dairy products have traditionally been judged according to traditional dairy defect standards, or evaluated by trained sensory panels. Results from these product assessment methods may then be paired with consumer acceptance data for extrapolating consumer insights from a sensory perspective. Thompson et al. (2004) showed that the combination of analytical and affective sensory techniques via preference mapping was successful in identifying drivers of liking for commercial chocolate milks for different consumer segments. Additionally, combinatorial sensory studies have been performed on unflavored fluid milk to understand drivers of liking, but only for specific populations (Chung, 2009), or for specific processing parameters (Gandy et al., 2008).

While sensory methods such as defect judging, trained panel profiling, and consumer acceptance testing are key in predicting consumer preferences, they explain only part of consumer behavior with commercial fluid milk offerings. Extrinsic product features that do not directly or explicitly affect the sensory qualities of a product, also play a large role in consumer purchase and acceptance. Willingness-to-pay and conjoint analysis studies have revealed that milk designated as rBST-free, organic, or local may justify significant price premiums commercially (Schott and Bernard, 2015; Harwood and Drake, 2018). Moreover, the presence of such features may serve to decrease perceived environmental or health risks (DuPuis, 2000) or satisfy alignment with altruistic motivations such as animal welfare (Costanigro et al., 2016; Harwood and Drake, 2018).

Overall, quantitative approaches such as consumer acceptance testing and surveys are exceptional tools for identifying key motivators behind purchase and acceptance. However, the nuanced belief systems and narratives behind those motivations are not easily ascertained with quantitative methods. For this reason, qualitative methods are often used to explain consumer insights from a more holistic context. Past fluid milk studies have primarily relied on focus group and one-on-one interviews to investigate motivators of acceptance or choice qualitatively. Hill and Lynchehaun (2002) used focus groups to investigate consumer-derived definitions for "organic" and "non-organic" and found them to be primarily tied to health beliefs, although actual knowledge of organic practices was substantially lacking among respondents.

Probing for gaps in education and finding anecdotal bases for belief systems are among the primary uses of in-person qualitative methods, but collection of such information is inherently at risk for introduced moderator bias (Tynan and Drayton, 1988). More recently, a structured approach for collection of qualitative data using assorted methods has been proposed to circumvent concerns of bias. In practice, this set of qualitative methods has been termed qualitative multivariate analysis (QMA), and consists of in-home usage testing, group discussions, product ranking, and projective mapping (PM) (Lopetcharat and Beckley, 2012). QMA methodology has been used to capture consumer insights for cottage cheese (Drake et al., 2009), cheese shreds (Speight et al., 2019), and pork products (Bittner et al., 2017); however, to our knowledge, QMA has not yet been utilized for understanding fluid milk consumer insights.

Regardless of what data collection method is used, understanding what product features influence acceptance and purchase can be difficult, as consumer actions are often at odds with stated opinions, beliefs, and preferences (Ulrich and Sarasin, 1995). For this reason, it is important to validate consumer insights by collecting complimentary data sources (qualitative vs. quantitative, implicit vs. explicit) so that congruency can be assessed and inconsistencies can be more aptly explained. Several studies have sought to join quantitative and qualitative data streams for explaining fluid milk consumer desires, however, these insights are generally limited to the combination of sensory analysis (CLT or DA) and focus group studies (McCarthy et al., 2017a; Porubcan and Vickers, 2005). Given the ubiquity of internet and computer access, large-scale surveys are increasingly used for garnering quantitative consumer insights. Unfortunately, application of surveys and the resulting collection of big data often fails to consider the diverse individual-level narratives behind the statistics. In order to bridge the gap between large-scale survey insights and individual-level qualitative insights, this study proposes to cross-validate conjoint-derived consumer segmentation with QMA for commercial fluid milks. Understanding the validity of insights derived from survey data on a qualitative level will provide a richer explanation for consumer purchase behavior, and the congruencies/incongruencies that are uncovered may provide aimed directions for educating and marketing to fluid milk consumers.

MATERIALS AND METHODS

Experimental Overview

An Adaptive Choice-Based Conjoint (ACBC) Analysis survey was conducted to understand consumer preferences for fluid milk offerings. Cluster analysis was subsequently performed on the estimated utility scores from the conjoint survey to understand segmentation among fluid milk consumers. Representative respondents from each identified consumer segment were then invited to participate in a four-week Qualitative Multivariate Analysis (QMA) study that included three weeks of home usage testing, followed by a focus group session in which participants discussed product experiences, ranked the products, and completed a projective mapping exercise. All testing procedures were conducted in compliance with North Carolina State University Institutional Review Board (NCSU IRB) regulations.

Adaptive Choice-Based Conjoint Analysis (ACBC)

Demographic questions, fluid milk usage questions, and an ACBC survey were developed using Lighthouse Studio (Sawtooth Software version 9.6.1, Orem, UT).
Conjoint analysis surveys, including ACBC, conceptually break products down into different features (attributes), and options within those features (levels). Randomly compiled product concepts can then be compared within the exercise in a format similar to real-life shopping situations. Repeated product comparisons allow for consumer choice patterns to be investigated, and multivariate analysis techniques can be subsequently applied to identify motivators of choice. While traditional CBC surveys would require analysis of all possible product combinations, the "adaptive" nature of ACBC allows for consumer input to refine what options are presented, resulting in lower sample size requirements and greater survey efficiency (Jervis et al., 2012).

Design and application of the ACBC survey was conducted as described by Harwood and Drake (2018). Briefly, the conjoint consisted of six attributes, each with two to five levels per attribute (Table 1). In addition, analysis of price and price premiums within the ACBC survey was conducted using the summed pricing technique (Cunningham et al., 2010). Several prohibitions were included within the conjoint design to ensure product concepts reflected real marketplace offerings for fluid milk. Prohibitions within the conjoint exercise included the following: rBST-free and organic, rBST-free and organic pasture-raised, shelf-stable and plastic jug. Participants were instructed prior to the conjoint exercise that organic milk offerings were, by definition, rBST-free, so the selection of both would be disallowed to ensure no doubling of price premium would be introduced. Following completion of the ACBC survey, respondents were asked if they were willing to participate in a 4-week in-person study on fluid milk that included in-home usage testing (weeks 1-3), as well as a focus group and projective mapping exercise (week 4) (QMA).

The ACBC survey was uploaded to the North Carolina State University Sensory Service Center database, which consists of over 12,000 members. In all, 719 respondents that self-reported purchase/consumption of fluid milk at least once per month completed the conjoint analysis survey. Participants who completed the entire survey were entered into a drawing to receive one of 15 $25 gift certificates to a local store.

Qualitative Multivariate Analysis (QMA)

In order to review the validity of segmentation from the conjoint analysis study, which documents consumer perception of product attributes without tangible products, fluid milk consumers from each of the four identified consumer segments were invited to participate in the QMA study. Selection for participation in the QMA portion of the study was determined based on root likelihood (RLH) values from the conjoint analysis survey, with respondents who had higher RLH scores receiving higher priority for inclusion. RLH is a measure akin to goodness of fit for discrete choice data. In all, 20 participants (5 from each segment) were chosen for the study, although only 18 (n=8 females and 10 males) completed the study. Participants who fully participated in the QMA study were compensated with a $100 gift certificate to a local store.

Nine commercial fluid milk products (all half-gallons, 1.5 weeks or more from expiration date) were chosen for the QMA study. These products were chosen to capture the breadth of features within the fluid milk market. Selected products included the following: Maple View Farms (locally-sourced, high temperature short time pasteurization (HTST)), Harris Teeter (store brand, HTST), Harris Teeter Organic (organic, store brand, ultrapasteurized (UP), Harris Teeter UP (store brand, UP), Organic Valley Omega-3 (national brand, organic, DHA-fortified, UP), A2 Milk (national brand, digestibility claim, UP), Fairlife (national brand, UP, ultrafiltered), Horizon Organic (national brand, organic, UP), and Organic Valley Grassmilk (national brand, organic, grass-fed). The home usage portion of the QMA study was conducted over a three-week period in a randomized complete block design, with each panelist receiving three of the nine possible milk products per week. Milks for each week of testing were picked up onsite at North Carolina State University and given to panelists in insulated bags with ice packs to ensure product quality and safety. Prior to participation, conjoint utility scores relating to milkfat preferences were evaluated. For participants whose utility scores suggested that they preferred milkfat-containing options (1%, 2%, or whole milk), 2% milk products were provided throughout the study. For participants who indicated that they preferred skim milk, skim milk products were provided throughout the study, with the exception of the Organic Valley Omega-3 and A2 Milk products, where 2% and 1% milkfat products were provided, respectively (no skim milk option was offered for these brands). Participants were instructed to try each milk at least once during the week, and were asked to access an online survey (Compusense Cloud, Guelph, Canada) where they rated their overall liking on a 9-pt hedonic scale and kept a diary of their likes and dislikes about the product following consumption. Participants were free to use the products in whatever way(s) they chose, so long as those actions mirrored their typical consumption patterns with fluid milk. Once poured or prepared, participants were asked to take a photograph of their chosen usage method and upload the photograph to their online journal entry for the given milk. After completing evaluations, panelists were required to wait a minimum of 10 h before either re-evaluating a product for a second time (optional), or moving on to their next sample.

Following the three weeks of home use evaluation of milks, participants shared their experiences and thoughts about the products during a single three-hour focus group session. Two focus group sessions (n=9 each session) were scheduled to accommodate panelist availability and to maintain a reasonable group size for inclusive discussion from all participants. The focus group sessions were video recorded and streamed externally to two note-takers to ensure key insights were captured. Prior to beginning the focus groups, consumers were provided with printed records of their home-usage testing diaries, uploaded photos of product use, and summaries of information such as name and price for each product. The first two hours of each session were oriented at understanding the values, beliefs, and desires that generally drive purchase, consumption and choice of fluid milk products. In addition, each product that was presented in the study was presented by the moderator and thoughts were gathered from each member of the focus group to understand how the aforementioned values and desires were met or unmet by certain products. The group then moved on to a short ranking exercise where they selected their favorite, second favorite, and least favorite of the milks.

Following a brief 15-minute break, the session finished with a projective mapping (PM) exercise. Panelists were given iPads with Compusense Cloud software projective mapping module and were asked to place the products on the map area according to product similarity/dissimilarity. During the PM exercise, panelists were able to review the product packaging, as well as their diary entries and notes from the focus group. Once the products had been placed in the map area, panelists had the option of tagging the products with an available list of descriptors. These descriptors included the following terms: low quality, high quality, artificial, natural, safe, expensive, affordable, misleading, sustainable, healthy, and animal welfare-conscious. Similar fluid milk descriptors were suggested to have high consumer relevance in observations reported by Cardoso et al. (2016). A field for free-response descriptors was also included.

Data Analysis

Responses from the ACBC survey were used to determine individual utility scores, importance scores, and RLH values through the application of hierarchical Bayesian (HB) regression (Sawtooth Software version 9.6.1, Orem, UT). Respondents with RLH values lower than 0.333 were subsequently removed from further data, as lower RLH values indicate a higher likelihood of "click-through" or random response patterns. In order to investigate the landscape of consumer opinions regarding fluid milk offerings in more depth, individual utility estimates (18 product feature levels + 7 stepwise price levels) from the conjoint survey were subjected to agglomerative hierarchical cluster (AHC) analysis (rows=719 respondents , columns= 25 total utility levels). AHC was performed using a dissimilarity matrix with Euclidean distance proximity type, Ward's agglomeration method, and entropy-based truncation for determination of the number of consumer segments (Cruz et al., 2013). Clustered conjoint data was subsequently subjected to a one-way analysis of variance (ANOVA) with Fisher's least significant difference post-hoc test (95% confidence).

Data from the PM exercise was exported and organized in a spreadsheet where rows (n=9) were assigned to each sample and columns (n=36) were arranged so that x and y-coordinates from each panelist's product placement were in sequential columns. In addition, counts of descriptor tag use were calculated for each sample and were arranged in a table as supplementary data columns (n=11). Multiple factor analysis (MFA) was subsequently applied to the coordinate data matrix, with the descriptor tag counts applied as supplementary variables to construct a biplot that visually explained product differentiation and associated descriptions (XLSTAT version 19.5.2018, Addinsoft, Paris, France).

RESULTS AND DISCUSSIONS

Conjoint Analysis Survey

Consumer segments from the conjoint survey were named according to insights gained from their conjoint utility and importance scores. As such, the clusters will be referred to throughout this study as the Premium, Value, Opportunistic, and Pragmatic clusters. Premium cluster members were defined by a higher utility for product features with high price premiums and suggestions of enhanced quality, as well as low care for price. Value cluster members were named as a result of high price sensitivity relative to other segments. Opportunistic cluster members were defined by a large range of suppressed price sensitivity, and a proclivity for choosing "premium" price features within their price comfort zones. Pragmatic cluster respondents exhibited features similar to both Opportunistic and Value cluster members, indicating a desire for certain target product features, but an overall adherence to high price sensitivity. These tendencies will be further explained in the following sections.

Demographically, consumer segments were similar in age and gender proportions, but were more distinct in breakdown of ethnicity, annual household income, and presence of children in the household (Table 3). Specifically, the Premium cluster was comprised of the highest proportion of non-Caucasian respondents (44.8%) and households with children (47.8%), as well as the lowest proportion of respondents who reported annual household income of

$100,000 or greater (22.4%). These demographics are consistent with previous literature which has documented a heightened likelihood for organic product purchase for non-Caucasian consumers, as well as an unclear link between annual household income and likelihood or organic/premium product purchase (Smith et al., 2009; Harwood and Drake, 2018). The value cluster, on the other hand, was comprised of the highest proportion of Caucasian respondents (81.4%), but was otherwise relatively distributed in terms of income and age. The pragmatic and opportunistic clusters were also relatively distributed demographically, although the pragmatic cluster had the lowest proportion of children in the household (31.4%) and the opportunistic cluster had the highest proportion of respondents from high-earning households (37.9% greater than $100,000/year).

Observations by Hill and Lynchbaum (2002) indicated that price is of primary importance for fluid milk consumers, even trumping features such as taste and quality. Similarly, price was of primary importance to each consumer segment in the present study; however, significant differences ($p<0.05$) in price importance were noted among the groups (Figure 1). Specifically, the Premium cluster placed the highest importance on non-price related milk features, suggesting that this group was most willing to pay price premiums for value-added features. The Opportunistic cluster was at parity ($p>0.05$) with the premium cluster in importance placed on fat content and milk type attributes, but placed a relatively low importance on label claims. The Pragmatic cluster was primarily price driven, but did receive an importance score for label claims that was significantly higher ($p<0.05$) than the opportunistic and value clusters. Finally, the Value cluster was lowest overall in allotted importance for non-price related attributes, indicating that fluid milk purchase for this group is focused exclusively on product cost.

Ideal builds were assessed based upon utility scores for each segment (Table 2). Some features, such as 2% milkfat and conventional pasteurization were unanimously preferred among all segments. However, consumer segments showed markedly different ideals for package type, milk type, and label claim preferences. For these attributes, the Premium cluster expressed preference for a fluid milk that was packaged in a plastic jug, organic pasture-raised, and had no label claims. The Pragmatic cluster showed similar preferences to the Premium cluster, with plastic jug packaging and no label claims; however, organic milk was the preferred milk type for the Pragmatic group. The Opportunistic cluster also had organic milk type in their ideal build, but additionally showed preference for cardboard carton packaging and rBST-free and DHA label claims. Finally, the Value cluster expressed a preference for conventional milk in a plastic jug, with no label claims, reinforcing the cluster's general preference for the most basic possible product. Utility for the "None" option, which indicates a group's utility threshold for purchasing a product (if summed utilities of product features from each attribute do not exceed this threshold, the consumer is likely to not choose no option, rather than the option being considered), was also assessed. The Value cluster, overall, exhibited the highest ($p<0.05$) "None" utility, indicating that they were the least likely to be satisfied by a given product offering, and most likely to walk away from a purchase decision if their needs were not met. The Premium and Pragmatic clusters exhibited a significantly lower "None" utility compared to the Value cluster, but significantly higher than the Opportunistic cluster, which was lowest overall. The relatively low "None" utility for the Opportunistic cluster indicates that this group is widely satisfied by various commercial milk offerings, and is more likely to buy a product that doesn't fit their ideals. Investigation into these patterns of preference and price sensitivity were of paramount concern in the subsequent QMA study to better define these concepts within each consumer segment.

QMA Study

Analysis of journal entries following in-home usage testing of commercial milks revealed that, overall, consumer opinions on the milks were driven most by price, flavor/aroma, and appearance. In terms of flavor, aroma, and mouthfeel, participants in the study noted several perceived differences among the commercial milks provided. While participants were unable to attribute these sensory differences to specific product features, previous studies on fluid milk have shown that both trained panelists and consumers are able to detect sensory differences in milk due to different pasteurization techniques (Lee et al., 2017), feed sources (Croissant et al., 2007), and package types (Simon and Hansen, 2001). Secondary features such as packaging type, milk type, nutritional information, shelf life, and functionality as an ingredient were also significant indicators of acceptability for some participants; however, package appearance and label claims (rBST-free, DHA-fortified, A2 protein), were more seldom mentioned. Although a variety of product features were mentioned, references to extrinsic product features such as milk type and label claim were generally discussed in terms of whether the given feature was "worth it" for the price of the product, reinforcing the baseline price sensitivity expressed in the conjoint exercise.

Cluster-related differences were also noted in the post-usage journal entries. Price sensitivity was relatively high for the Value, Opportunistic, and Pragmatic clusters, with references to price or affordability appearing in 69.8% (88/126) of journal entries, compared to only 27.8% (10/36) in the Premium cluster. This reflects the relatively low importance that the Premium cluster exhibited for price in the conjoint study, but suggests that the decreased importance is not simply a function of seeking premium features. Rather, it appears that price is simply not a feature that is considered as often in the decision-making actions for this group in regard to fluid milk. Furthermore, annual household income and household sizes reported by this group suggests that they have no clear difference in disposable income compared to other groups, further reinforcing an innate difference in approach when assessing commercial milk purchase. Among other differentiating trends found in the journal entries, the Pragmatic cluster expressed significantly more concern than the other groups for shelf-life and nutritional features. While no clear inclination for shelf-life importance was noted in the conjoint study, the Pragmatic cluster did place significantly more importance on label claims than the Opportunistic or Value clusters, which may show that this group is more likely to read and explore product information that is readily available on the package to ensure they make an informed purchase. In this way, attention to shelf life by Pragmatic cluster-type consumers may be driven by a desire to utilize the product features they sought out without fear of spoilage.

Photo uploads from the QMA diaries revealed that commercial milks were consumed in a variety of ways. Primarily, the milks were consumed, in order of frequency, in the following ways: as-is, with cereal, as a coffee creamer, as a meal ingredient, and as base for shakes/smoothies. Preference for usage style was relatively consistent throughout the study, with most consumers opting to consume all nine milks in the same fashion. The Pragmatic and Opportunistic clusters were the least likely to consume the milks on their own, choosing instead to utilize the milks as functional ingredients (ex: frothed for coffee) or flavor compliments (ex: base for smoothie). In a review of milk proteins, Fox (2001) noted that adult populations primarily consume milk as an ingredient, and, as such, may derive value from milk's functional properties. As noted in the conjoint study, the Opportunistic and Pragmatic clusters were defined by choice patterns that seemingly keyed in on specific product features. These choice patterns, coupled with differences in propensity to review product information and consume milk products in a variety of fashions, imply that purchase behaviors may be governed by seeking features they feel imply quality or functionality for a given use. Furthermore, the Opportunistic cluster exhibited a relatively high utility for ultrapasteurization, although conventional pasteurization was their ideal option. While this result may simply reflect a lack of knowledge of commercial organic milk production (typically ultrapasteurized), it may also suggest that Opportunistic consumers seek a product with a longer shelf life because their uses for milk are outside of simply drinking it as a beverage.

Focus groups with QMA participants were oriented at more thoroughly understanding consumer experiences with the tested commercial milks, as well assessing the value of various product features. Furthermore, these opinions were used to add context to the analysis of the projective mapping exercise and home usage test results. Visually, product spaces from the projective mapping exercise indicated that there was clear differentiation of the commercial milks (Figure 2). Panelists tended to differentiate the milks based on perception of quality and affordability on Factor 1 (56.01% variability explained) and based on trust factors (misleading, artificial, safe, natural) on Factor 2 (21.58% variability explained). Non-organic store brand products (Harris Teeter, HTST and UP) were generally associated with being affordable, but low quality. The concept of private label milks being perceived as lower quality has been well-documented in previous studies, although price and store image may significantly affect these perceptions of private label grocery products (Hovhannisyan and Gould, 2012; Richardson et al., 1994). Liking scores (not pictured) for these products indicated that the Harris Teeter UP product was among the bottom three milks in liking for each group, except the Value cluster, for which it was tied for the highest liking score among the milks evaluated.

Milks such as A2 Milk, Horizon Organic, and Organic Valley Grassmilk were associated with being expensive and high quality, as well as healthy, sustainable, animal welfare-conscious, safe, and natural. Overall, each of these milks received relatively high liking scores from each consumer group, with no milk receiving a bottom three score in liking; however, the highest liking scores came from participants in the Pragmatic and Opportunistic clusters. Specifically, the Pragmatic cluster gave its highest liking scores to Horizon Organic and A2 milk. Preference for a national brand organic product reflected conjoint preferences for the Pragmatic group, whereas the acceptance for A2 milk highlighted the group's aforementioned tendency to review product details and make an informed choice to meet their needs. McCarthy et al. (2017b) showed that both milk and plant-based milk alternative consumers value products that promise digestive benefits. While no clear within-cluster trend was determined during the focus groups, many participants expressed interest in the A2 product and claims for better digestibility, suggesting further that this may be a motivating feature for milk consumers. The Opportunistic cluster expressed a similarly high opinion for the Horizon Organic product, scoring it as their second most-liked sample, which mirrored conjoint preferences for a product that was organic and packaged in a cardboard carton. Organic Valley Grassmilk was the most liked sample for the Opportunistic cluster, overall. While the most preferred milk type option in the conjoint study was organic for the Opportunistic cluster, they did receive the highest utility score ($p<0.05$) for the organic pasture-raised milk type among all clusters. This suggests that consumers within this segment are characterized by a baseline desire for organic designation in their commercial milk choice and are willing to explore additional "premium" features or claims if the total product price is within their price comfort range. Analysis of price utility scores for the Opportunistic cluster suggests that, compared to other groups, they have a relatively large price range they would consider for commercial milks. While most groups exhibited a proportional decrease in utility with increase in price, the Opportunistic group showed stable utility scores until a total price of about $5.00 was reached, after which utility steeply dropped (Figure 3). Given the $5.00 price point coincides with the price threshold of several "premium" commercial milk offerings, it is unsurprising that the Opportunistic cluster is defined by preference for these items, but are willing to explore more basic products if the premium products are too expensive.

Along Factor 2, the descriptors "artificial" and "misleading" loaded positively, and were strongly associated with the Fairlife and Organic Valley DHA milk samples. Focus group insights indicated that many consumers believed these samples to be "artificial" for different reasons. For the Fairlife sample, several participants pointed out that the removal of lactose and difference in protein content as a result of ultrafiltration made the product, by definition, artificial in nature, although many also reported these features as a value-add. Furthermore, many noted in the focus group and in their journals that the Fairlife sample was markedly different in aroma and mouthfeel compared to the other milks. Overall, both the Fairlife and Organic Valley DHA samples received polarized liking scores among the consumer clusters. The Pragmatic cluster gave a higher score to Fairlife than any other group, indicating that high protein content or lactose-free might be motivating features for a subset of fluid milk consumers. Similarly, the Organic Valley DHA milk was liked strongly by the Premium cluster and, surprisingly, the Value cluster, whereas it was least liked for the Pragmatic cluster and second-least liked for the Opportunistic cluster.

In contrast to the Fairlife and Organic Valley DHA milks, the Harris Teeter Organic and Maple View Farms (Hillsborough, NC) milks were negatively correlated with “artificial” and “misleading” descriptors. Additionally, these products did not load along Factor 1, indicating that these samples were, overall, believed to be intermediate in terms of affordability and quality. The Harris Teeter Organic product did not elicit strong reactions from any group, generally scoring towards the middle for each (between 3rd and 6th). Journal responses and focus group notes echoed the implications of these intermediate liking scores, with more price-sensitive panelists (Value/Opportunistic/Pragmatic cluster members) indicating price was too high, and others commenting that the milk was “no real standout” (Pragmatic cluster member) and many other milks were “better than this one” (Premium cluster member). The Harris Teeter Organic product was also particularly criticized in journal entries as having a strange flavor and aftertaste, which several focus group participants attributed to ultrapasteurization. Decreased liking resulting from cooked flavors associated with ultrapasteurization have been widely noted in previous milk studies (Gandy et al., 2008; Lee et al., 2017), although there was no clear indication of why other ultrapasteurized samples in the study were not penalized. On the other hand, the Maple View Farms (local) milk was more polarizing among the consumer groups.

The locally-farmed milk (Maple View Farms) was not very appealing for members of the Pragmatic and Opportunistic clusters (rated 7th and 4th, respectively); however, it particularly appealed to Premium and Value cluster consumers, scoring 1st for each group. Premium cluster members spoke highly of the milk quality (flavor, aroma, mouthfeel), as well as the local-designation, which was a perceived as a value-added feature compared to other milks in the study. In addition, Premium cluster members reported that the glass container “made [the milk] seem fresher” and induced a nostalgic feeling that represented “the old way milk was bought”, corroborating focus group insights on glass milk packaging reported by Hollywood et al. (2013). Value cluster members found the Maple View Farms milk appealing for many of the same reasons as the Premium cluster: sensory quality, attractive container, and local-designation. In the conjoint exercise, the Value cluster expressed a preference for conventional milk, likely due to the lower price; however, locally-farmed milk type designation was the next most preferred milk type for the group, and was the highest average utility for locally-farmed among all groups. In the case of Maple View Farms, Value cluster members also reported that the milk was a desirable option because the milk had a bottle-buy-back program that would credit them $1.50 for returning the bottle to their grocer. In this way, the Value cluster effectively mirrored their conjoint-derived preference for local designation, while maintaining their relatively strict price standards. Other QMA participants mentioned that the bottle deposit system was a feature they were unlikely to utilize, thereby leaving their perception of price unchanged for this product. Observations by Grimes-Casey et al. (2007) suggest that milk consumers are, overall, more likely to participate in deposit-refund programs for milk if the deposit is high, although their initial purchase likelihood decreases proportionally to the increased deposit price. Taking into account the draw that local designation and glass packaging presents for some fluid milk consumers, investigation of ideal deposit-return pricing models that work for both consumers and retailers may be warranted.

CONCLUSIONS

Cross-validation of conjoint analysis and QMA studies revealed that survey-derived consumer sentiments for fluid milk largely agreed with qualitative measures such as home usage testing and focus groups. Generally, this suggests that rapid testing procedures, such as conjoint analysis, are able to accurately predict consumer segmentation for commodity products such as fluid milk. As indicated in the conjoint study, price sensitivity was the largest determinant of consumer segmentation, although certain consumer groups did value other “premium” product features. Beyond price, consumer clusters in the study were defined by their unique preferences for milk type, packaging, and the presence/absence of other value-added features. Specifically, the Premium cluster exhibited preferences for non-conventional milk types that imply higher quality. The Value cluster valued the most basic available product features, but showed a willingness to explore locally-farmed milks. The Pragmatic cluster allotted relatively little importance to anything but price, but decisively preferred organic milk to other available milk types, including pasture-raised and organic pasture-raised, which are often conflated with organic designation. Finally, the Opportunistic cluster expressed acceptability for a larger group of product options compared to other groups, but generally preferred organic milk packaged in cardboard/paperboard cartons with added features such as DHA-fortification or rBST-free label claims. The application of QMA showed that these preferences generally play out in real-life interactions with commercial milk offerings, and may additionally be influenced by factors such as household size, personal beliefs, convenience, and proclivity to read product information/labeling. Additionally, QMA insights and application of projective mapping suggested that consumers implicitly relate non-conventional milk types with higher quality. To this end, investigation into the presence of implicit biases related to milk production and milk type designation is a needed area of future research. Furthermore, QMA revealed that consumers perceive differences in flavor, aroma, and mouthfeel among commercial milk products. While these features may not be readily captured using online survey techniques such as conjoint, they may help expand on consumer likelihood of repeat purchases, assuming the conceptual needs required for initial purchase are met. Understanding of consumer insights for fluid milk from a macro survey perspective, supported by anecdotal qualitative insights is extremely valuable for developing targeted marketing strategies and is integral for future growth within the fluid milk market.

ACKNOWLEDGEMENTS

This study was funded in part by The Dairy Alliance (Atlanta, GA) and the National Dairy Council (Rosemont, IL). The use of trade names does not imply endorsement or lack of endorsement of those not mentioned.

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Table 1. Attributes and levels used in fluid milk conjoint survey

Fat content	Milk type	Package type	Shelf life[1]	Label claims[2]	Price
Skim	Conventional	Plastic jug	Conventional pasteurization (HTST; ~18-21 d)	None	Summed pricing[4]
1% Milkfat	Organic ($2.37)	Cardboard carton	Ultrapasteurized (~30-65 d)	rBST-free ($0.24)	
2% Milkfat	Locally farmed ($1.00)		Shelf-stable (does not have to be refrigerated until opened	DHA-fortified[3] ($0.48)	
Whole	Pasture-raised ($1.76)			rBST-free[3] and DHA-fortified ($0.72)	
	Organic pasture-raised ($4.14)				

[1] Harwood and Drake (2018)

[2] Definitions were explained in terms of refrigerated shelf life

[3] DHA= docosahexaenoic acid; rBST= recombinant bovine somatotropin

[4] Summed pricing refers to additive pricing for select product features that carry a price premium. Total summed price (± 30%) was shown for each assembled product concept in the conjoint exercise.

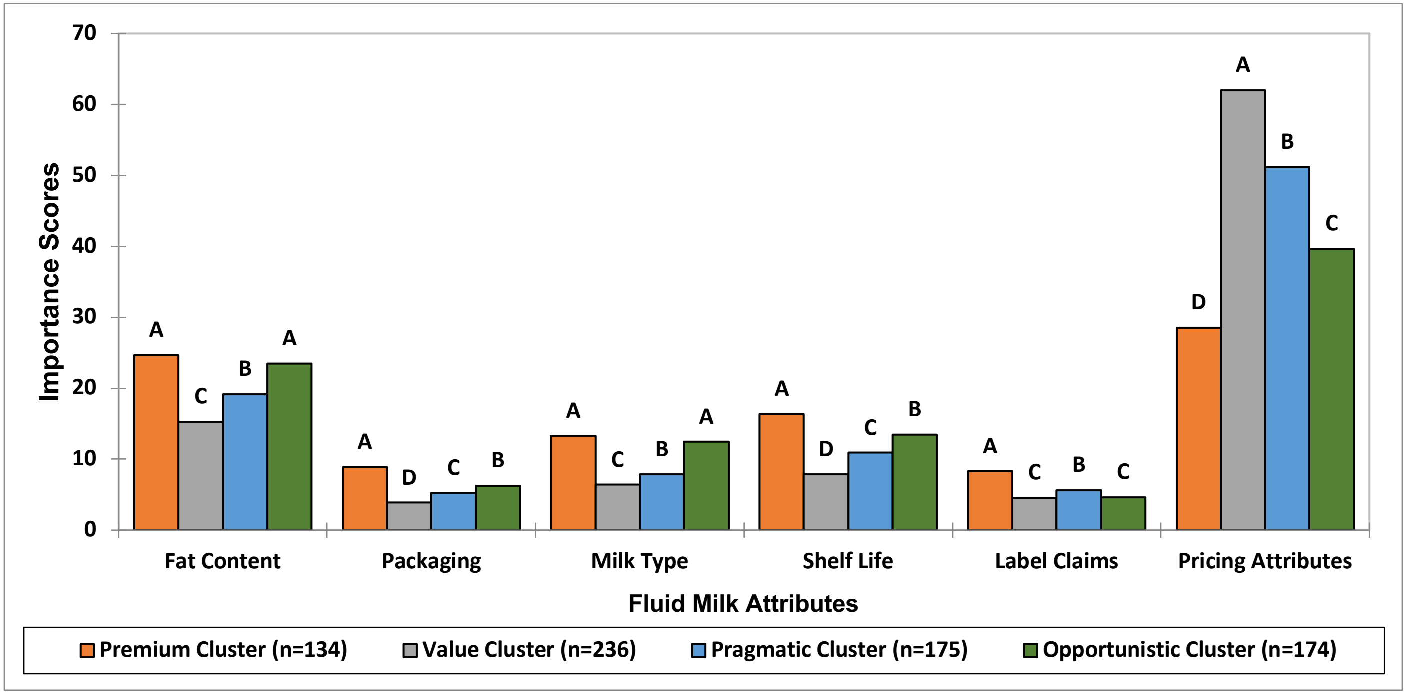

Figure 1. Attribute importance scores from fluid milk conjoint survey for segmented consumer clusters. Letters within attributes (a-d) indicate significant differences between clusters ($p<0.05$).
*Sum of importance score values for each cluster is 100 points total and scores are interpreted as ratio-scaled values.

Table 2. Average utility scores from fluid milk conjoint survey for segmented consumer clusters

		Premium Cluster (n=134)	**Value Cluster (n=236)**	**Pragmatic Cluster (n=175)**	**Opportunistic Cluster (n=174)**
Fat Content	1%	-2.6b	10.3a	10.0a	-3.6b
	2%	***37.7a***	***14.8b***	***20.9b***	***34.3a***
	Whole	1.5b	-17.6c	-19.0c	26.7a
	Skim	-36.6b	-7.6a	-11.9a	-57.4c
Package Type	Cardboard Carton	-6.1c	-4.8bc	-1.1b	***10.7a***
	Plastic jug	**6.1a**	***4.8ab***	***1.1b***	-10.7c
Milk Type	Conventional (non-organic)	-9.0c	***12.2a***	0.9b	-32.7d
	Organic	8.6b	-2.9c	***5.4b***	***26.3a***
	Locally farmed	-0.2b	4.9a	2.2ab	0.7b
	Pasture-raised	-8.6c	-1.4a	-4.6b	-6.7bc
	Organic Pasture-raised	***9.2a***	-12.8c	-3.9b	12.4a
Shelf Life	Conventional pasteurization (HTST; ~18-21 d)	***43.2a***	***19.2c***	***25.6b***	***29.1b***
	Ultrapasteurized (~30-65 days)	-0.1b	2.6b	2.4b	10.7a
	Shelf-stable (does not have to be refrigerated until opened)	-43.1c	-21.7a	-28.0b	-39.7c
Label Claims	None	***10.2a***	***11.7a***	***10.9a***	2.0b
	rBST-free	-5.3b	-5.9b	-6.4b	-1.5a
	DHA-fortified	-1.5a	-6.3b	-4.1ab	-3.5a
	rBST-free and DHA-fortified	-3.4c	0.4b	-0.5b	***3.0a***
Overall None Utility		18.9b	33.6a	18.6b	-10.5c

*Bolded items within columns indicate ideal build features for that given cluster

**Means in a row followed by a different letter are different ($p<0.05$)

Table 3. Demographic information for fluid milk consumer clusters (n=719)

		Premium Cluster (n=134)	**Value Cluster (n=236)**	**Pragmatic Cluster (n=175)**	**Opportunistic Cluster (n=174)**
Gender	Male	30.6%a	26.7%a	22.9%a	24.1%a
	Female	69.4%a	73.3%a	77.1%a	75.9%a
Age	18-25	22.4%ab	26.7%a	26.9%a	14.4%b
	26-35	28.4%a	21.2%a	28.0%a	32.2%a
	36-45	23.9%a	21.6%a	17.7%a	26.4%a
	46-55	16.4%a	16.1%a	13.1%a	14.9%a
	56-65	6.0%a	11.9%a	9.7%a	9.2%a
	66 and older	3.0%a	2.5%a	4.6%a	2.9%a
Ethnicity	White	55.2%c	81.4%a	69.1%bc	70.7%ab
	Black/African American	25.4%a	6.8%b	14.9%ab	12.6%b
	Latino or Hispanic	3.0%a	5.9%a	1.7%a	4.0%a
	East Asian	3.0%a	4.2%a	5.7%a	5.7%a
	South Asian or Indian	10.4%a	2.5%a	8.6%a	6.3%a
	Middle Eastern	0.7%a	0.8%a	0.6%a	0.0%a
	Native American	0.7%a	0.8%a	0.0%a	0.6%a
	Other	3.0%a	0.8%a	1.1%a	1.7%a
	Prefer not to answer	3.0%a	1.3%a	0.6%a	2.3%a
Annual Household Income	Under $25,000	6.0%a	8.5%a	8.0%a	5.2%a
	$25,000 - $49,999	22.4%a	20.8%a	17.7%a	17.2%a
	$50,000 - $74,999	42.5%a	33.9%a	41.1%a	32.8%a
	$100,000 or greater	22.4%b	28.8%ab	27.4%ab	37.9%a
	Prefer not to answer	6.7%a	8.1%a	5.7%a	6.9%a
Number in Household	1	13.4%a	16.5%a	11.4%a	14.9%a
	2	35.1%a	33.9%a	40.0%a	37.4%a
	3 or more	51.5%a	49.6%a	48.6%a	47.7%a
Children in Household	No	52.2%b	65.3%ab	68.6%a	59.2%ab
	Yes	47.8%a	34.7%ab	31.4%b	40.8%ab
Regular Grocery Shopping Locations*	Premium Grocery Stores	61.2%a	37.3%b	53.7%a	63.2%a
	Bulk supplier	51.5%a	50.4%a	44.6%a	50.0%a
	Discount Grocery Stores	77.6%a	84.7%a	85.7%a	75.9%a
	Standard Grocery Stores	95.5%a	96.6%a	97.7%a	96.0%a
	Farmers Markets	37.3%ab	25.8%b	31.4%ab	43.7%a
	Other	1.5%b	5.5%ab	5.1%ab	8.6%a

*Check-all-that-apply question. Totals may exceed 100%.

**Percentages in a row followed by a different letter are different ($p<0.05$).

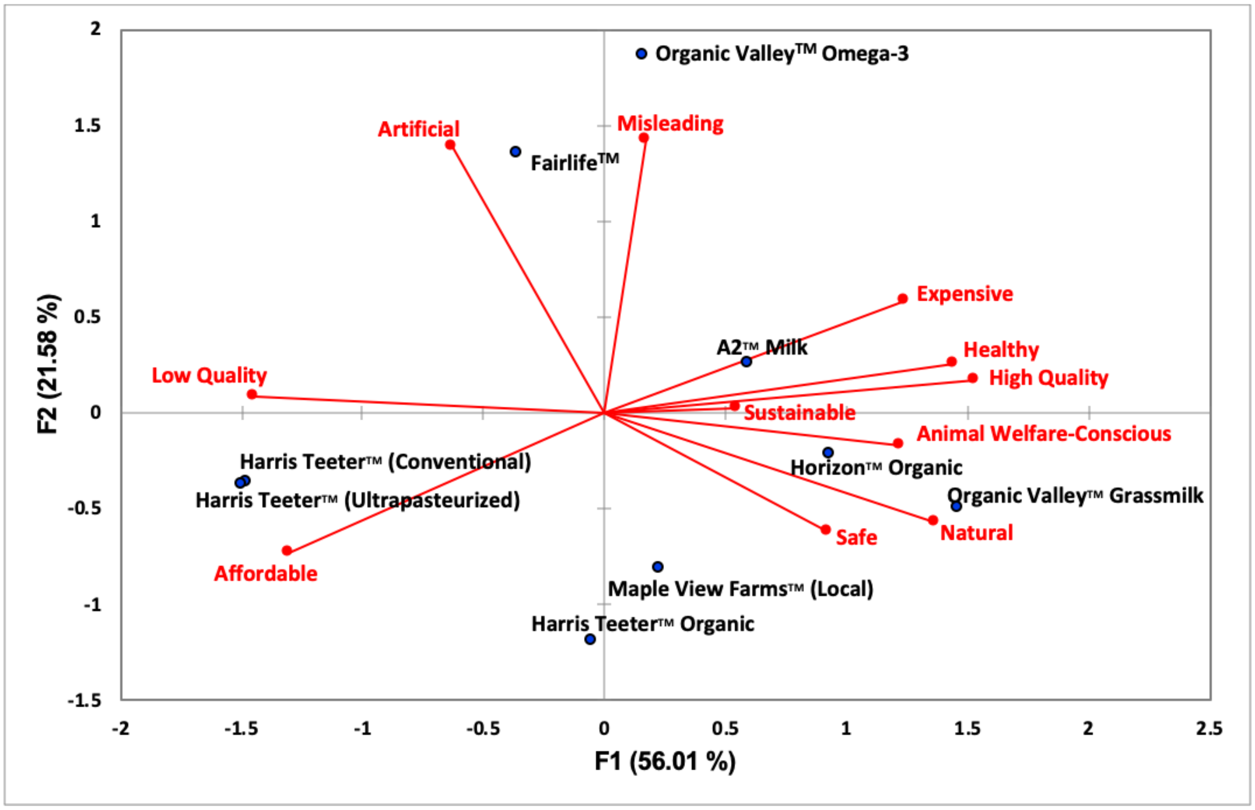

Figure 2. Projective map of commercial brand fluid milks and descriptive product features from QMA study participants (n=18)

Figure 3. Conjoint-derived price sensitivity of fluid milk consumer clusters

CHAPTER 3: The Role of Heat Treatment on Light Oxidation of Fluid Milk

The Role of Heat Treatment on Light Oxidation of Fluid Milk

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The content of this chapter was published in:

Journal of Dairy Science (in press)

ABSTRACT

Light oxidized flavor (LOF) resulting from photooxidation of riboflavin following light exposure is one of the most common off-flavors in fluid milk. The sensory perception of LOF has been studied extensively in high temperature short time pasteurized (HTST) milk, but few studies have evaluated ultrapasteurized (UP) milk. The objective of this study was to evaluate the role of heat treatment on the development of LOF in UP fluid skim milk. Skim milk was processed by HTST or by direct steam injection (DSI-UP) and subsequently exposed to 2000 lx light emitting diode (LED) light for various times. Sensory properties were monitored by descriptive analysis and threshold tests, while volatile compounds were evaluated by solid phase microextraction (SPME) with gas chromatography mass spectrometry (GCMS). Dissolved oxygen and riboflavin were determined at each time point by an oxygen meter and ultra-performance liquid chromatography with a fluorescence detector (UPLC-FLR), respectively. The entire experiment was performed in triplicate. Typical cardboard and mushroom flavors (LOF) were detected by trained panelists in HTST milk after 3.5 h of light exposure. In contrast, LOF was not detected by trained panelists in UP milk until 36 h of light exposure. Similarly, the best estimate threshold for LOF from untrained consumers (n=101) was higher ($p<0.05$) for DSI-UP milk (61.0 h) than HTST milk (15.2 h). Milks with LOF were characterized by higher ($p<0.05$) relative abundance of lipid oxidation compounds hexanal and heptanal. Dissolved oxygen (DO) and riboflavin concentrations decreased ($p<0.05$) with increased light exposure time and the decrease was slower ($p<0.05$) in UP milk compared to HTST milk. Initial DO concentration was investigated as a possible influence in LOF development because DSI-UP milks had lower ($p<0.05$) initial DO concentrations than HTST milks. However, follow-up evaluations of deaerated HTST milks suggested that DO was not a significant factor in LOF development.

These results demonstrate that UP milk is less sensitive to LOF than HTST milk, possibly due to sensory masking effects or antioxidant effects of volatile sulfur compounds. An enhanced understanding of light and storage effects on milks will assist with best practices when transporting and displaying fluid milk products for sale.

INTERPRETIVE SUMMARY

Light oxidized flavor (LOF) in fluid milk is a quality issue that can lead to decreased consumer acceptance. This study found that ultrapasteurized (UP) skim milk developed light oxidized flavors slower than high temperature short time (HTST) skim milk following light exposure, as evidenced by both consumer and trained sensory panel evaluations. Furthermore, instrumental analyses suggested that the observed delay in LOF development could be attributed to volatile sulfur compounds which likely played a synergistic masking and antioxidant role.

Keywords: milk, oxidation, pasteurization, threshold

INTRODUCTION

Thermal processing of fluid milk is central to fluid milk quality. Under typical high-temperature-short-time (HTST) pasteurization conditions (minimum of 72°C for 15 s), fluid milk has a shelf life of approximately 3 weeks, after which milk quality deteriorates due to microbial load. Shelf life of fluid milk may be significantly extended through the application of elevated thermal processing. Ultrapasteurization (UP) of fluid milk (minimum of 138°C for at least 2 s), for example, results in a shelf life of about 60 days (Boor and Nakimbugwe, 1998). Ultrapasteurized fluid milk is advantageous to fluid milk producers and retailers because the extended shelf life of such products reduces risk of product loss and helps assuage supply chain demands. However, elevated pasteurization temperatures have been shown to impart increased cooked and sulfur flavors, which result in decreased consumer liking (Chapman and Boor, 2001; Gandy et al., 2008). Lee et al. (2017) showed that milk consumers, both adults and children, maintained preference for HTST milk over UP milk regardless of fat content. Additionally, Lee et al. (2017) demonstrated that the cooked and sulfur/eggy flavors imparted by UP treatment of fluid milk decreased over shelf life, but were still distinct through 14 d post-processing.

While extended shelf life is an attractive feature to fluid milk processors and retailers, longer time in a retail setting increases the likelihood of extended light exposure. The development of off-flavors in milk due to photooxidation of riboflavin and other photosensitive compounds in milk has been widely linked to increased concentrations of volatile aldehydes such as methional, hexanal, heptanal, and octanal, and a corresponding decrease in consumer acceptance (Johnson et al., 2015; Brotherson et al., 2016). Aldehyde formation in fluid milk is primarily caused by light-induced degradation of methionine and oxidation of unsaturated fatty acids (Marsili 1999; Min and Boff, 2002). Accumulation of volatile aldehydes and other oxidation-related compounds results in off-flavor development, commonly referred to as light activated flavor or light oxidized flavor (LOF). In skim fluid milk and other dairy products, LOF is commonly described as “burnt”, “tallow”, “cardboard”, “mushroom”, or “cabbage-like”, although several other descriptors have been used (Bodyfelt et al., 1988; Brotherson et al., 2016).

Mitigation of light oxidation has been most commonly addressed through the use of light-blocking packaging materials. Traditionally, UP milk has been packaged in gable-top polyethylene-coated paperboard containers, which provides extensive, yet imperfect protection from light (Rysstad et al., 1998). Single-serve containers and larger volume polyethylene packages have also emerged for UP milk products. Paperboard cartons provide strong light protection, but the majority of fluid milk consumers have reported a preference for packaging materials that allow for product visibility, such as glass, high-density polyethylene (HDPE), or polyethylene terephthalate (PET) (Wang et al., 2018). Mestadgh et al. (2005) showed that HDPE bottles with either an added oxygen-binding inner layer or ultraviolet (UV) light filter failed to stop light oxidation in UP milk; however, the addition of a UV-absorbing material offered slight protection. Use of TiO_2 pigments, or secondary light-blocking materials, such as foil wraps, were shown to be successful in mitigating light-oxidation in reduced-fat UP milk (Stancik et al., 2017); however, these applications may present significant cost barriers (compared to standard HDPE containers) and may be negatively perceived by consumers in shopping situations due to decreased product visibility (Webster et al., 2009; Wang et al., 2018).

In addition, the efficacy of antioxidant addition on the prevention of light oxidation in fluid milk has been investigated. Experiments by van Aardt et al. (2005) reported that addition of α-tocopherol decreased hexanal formation during storage, while additions of butylated hydroxyanisole and butylated hydroxytoluene decreased pentanal, hexanal, heptanal, and 1-

octen-3-ol, although consumer-relevant intensities of LOF were not assessed. The addition of vitamin premixes were found to have no effect on LOF development in HTST skim milk (Schiano et al., 2019). Flavor and sweetener addition, although not directly oriented at the mitigation of oxidation, has also proven effective in protecting fluid milk from the negative sensory effects of LOF development. Chapman et al. (1998) determined that the addition of cocoa and carrageenan to fluid milk successfully inhibited vitamin A degradation, but was ultimately ineffective in preventing LOF development. While flavor addition failed to prevent LOF, threshold studies by van Aardt et al. (2001) showed that consumers of whole (3.25% milkfat) chocolate milk had a significantly higher threshold for light-induced off-flavor than for unflavored fluid milks (of various fat levels). Similar findings were also reported in fluid milks formulated with sucrose and strawberry flavor (Bills et al., 1972). Results from these studies indicate that flavored milk formulations may not directly address LOF development, but flavoring can serve as a means for masking off-flavors and preserving consumer acceptance.

Intrinsic qualities of fluid milk such as milkfat content also influence oxidative stability and development of LOF. In terms of milkfat, Whited et al. (2002) showed that under 2000 lx fluorescent light, skim milk had greater vitamin and riboflavin loss than reduced fat or whole milk and LOF developed more rapidly at early timepoints (2h light exposure); however, LOF intensity was greater in reduced fat and whole milk samples, compared to skim milk, after extended light exposure (4h or more). While greater milkfat content may result in higher intensities of LOF, consumer evaluations indicate that overall liking of 2% milk was penalized less than skim milk following light exposure, in agreement with general consumer preference for milk with higher fat content (Martin et al., 2016; McCarthy et al., 2017). Skim milk, as such, remains the most flavor-transparent matrix for evaluation of LOF in milk.

Overall, there has been limited investigation into the roles different processing parameters play on the development of light oxidation in fluid milk. Existing literature primarily addresses the phenomenon indirectly through evaluation of riboflavin degradation. Saidi and Warthesen (1995) reported that heat treatment (80°C, 100°C, and 120°C) and homogenization (single stage- 35 kg cm^{-2}; double stage- 175 kg cm^{-2} and 35 kg cm^{-2}) of skim milk resulted in decreased rates of riboflavin degradation, although this improvement was primarily attributed to greater light scattering following homogenization. The objectives of this study were to investigate the relationship between thermal processing and development of LOF in skim milk using both analytical measurements (descriptive analysis, volatile flavor analysis, riboflavin measurement), as well as consumer-relevant methodologies (threshold testing). A holistic understanding of these processes may help guide best practices related to processing, storage, and display of commercial fluid milks to ensure ideal product quality.

MATERIALS AND METHODS

Milk Processing and Light Exposure

Raw skim milk (200 L per lot, 0.083% ± 0.001% fat) for this study was obtained from the North Carolina State University Dairy Enterprise System (Raleigh, NC). Somatic cell count (SomaScope, Delta Instruments B.V., Drachten, the Netherlands) of the raw milk was <100,000 cells/ml for each lot of milk, and each lot had <10 cfu/ml coliforms. Each lot of skim was divided into two portions (100 L each) and high-temperature-short-time pasteurization (HTST) or direct steam injection (DSI) ultrapasteurization (UP) were conducted using a MicroThermics EHVH pasteurization unit (MicroThermics, Raleigh, NC) with a 2-stage homogenizer (GEA Niro Soavi, Parma, Italy) as described by Lee et al. (2017) (Figure 1). Microbial counts and alkaline phosphatase tests were conducted post-pasteurization as described by Lee et al. (2017).

Following thermal processing and cooling to 10°C, milks were bottled in translucent, half-gallon, high-density polyethylene milk jugs (Upstate Niagara Cooperative Inc., Buffalo, NY) under an Aseptilab™ ultra-clean fill hood (MicroThermics, Raleigh, NC). Milk containers were placed in a walk-in cooler and stored in the dark at 4°C for 48 h to simulate post-process transit time, similar to commercial products. These processes were repeated in triplicate.

Following 48 h at 4°C, milks were placed into one of two light boxes (88 x 57 x 31 cm) constructed as described by Schiano et al. (2019). Light boxes were covered with laser cloth (BK5 blackout fabric, ThorLabs Inc., Newton, NJ) to eliminate ambient light from reaching samples during exposure. Within each light box, light-emitting diode (LED) strips (3500K, International Light Technologies, Peabody, MA) were affixed to the roof to supply a light intensity of 2,000 lx (+/- 20 lx) throughout the box at the height of the milk jugs. Light boxes were located within a walk-in cooler at 4°C.

Experimental exposure times for HTST and DSI-UP milks was determined following a preliminary tasting trial with untrained assessors (n=16) from the general North Carolina State University student population. Milks from each treatment were exposed to light at 4 h increments, for up to 48 h. Trial participants were asked to indicate at what point they believed a noticeable difference was imparted to the milk samples by first tasting an unexposed control sample, and then tasting the exposed milks in order of exposure time. Participants were presented with treatments in a balanced design to mitigate order effects. Following these trials, suggested midpoints of 8 h and 36 h were chosen for HTST and DSI-UP milks, respectively. A 1.5x step factor was subsequently applied to the midpoint of each treatment, resulting in the following experimental time points for the study: HTST samples were removed from the light boxes at 2.4 3.6, 5.3, 8, 12, 18, and 27 h and DSI-UP milks, samples were removed at 10.67, 16, 24, 36, 54, 81, and 121.5 h. Once removed, milks were placed in a light-shield box covered with laser cloth to ensure no further light exposure was introduced. Unexposed control milks for each pasteurization method were also prepared and placed directly into a light-shielded box post-processing. At the conclusion of the 121.5 h maximum exposure time, all milks were subjected to descriptive sensory analysis, sensory threshold testing, and instrumental analyses. The entire experimental design was replicated in triplicate.

Riboflavin and Dissolved Oxygen Measurement

Riboflavin content and degradation over the course of light exposure was determined in accordance with the procedure developed by Silva et al. (2005). Sample preparation consisted of protein precipitation using tricholoroacetic acid, centrifugation, and filtration of the supernatant through a 0.2 µm nylon filter. Riboflavin concentration was measured by ultra-performance liquid chromatography (UPLC; Acquity H-Class; Waters Corporation, Milford, MA) with fluorescence (FLR) detection (excitation = 420 nm, emission = 530 nm). The concentration of riboflavin (µg/mL) was calculated using a relative response factor determined with calibration standards (0.1-0.3 µg/mL) (Sigma Aldrich). Dissolved oxygen (DO) was measured using an YSI-5100 dissolved oxygen meter with a self-stirring probe (YSI Inc., Yellow Springs, OH, USA). The instrument was calibrated prior to each use using a biochemical oxygen demand (BOD) bottle containing 1” water, in accordance with the device manual. Milk aliquots (400ml, 10°C at time of measurement) from two bottles of each thermal process/light exposure treatment were prepared and measured for each replication of the experiment by submerging the self-stirring probe in the milk aliquot.

Trained Panel Profiling

Sensory analysis of milks was conducted to understand the relevant sensory changes brought about from light exposure. Sensory profiling of milks was approved by the NCSU Institutional Review Board for Human Subjects. A trained panel of eight descriptive analysis (DA) panelists (2 males, 6 females; ages 21-55 y), each with at least 150 h of prior experience profiling fluid milk and dairy products, profiled intensities of typical milk flavor attributes (Jo et al., 2018) and light oxidation-specific flavors such as cardboard and mushroom (Schiano et al., 2019) using the Spectrum™ method 15-point intensity scale (Meilgaard et al., 2007) (Table 1). Forty-ml aliquots of milk from each treatment were dispensed into lidded 65 ml soufflé cups labeled with random three-digit blinding codes and were subsequently lidded and equilibrated to 15°C. Trained panel profiling was conducted in a room with overhead lights turned off to ensure no further light exposure was introduced to the samples. Data from each panelist was collected on an iPad interface with Compusense Cloud (Guelph, Ontario) software. Each milk from each experimental replicate was evaluated in duplicate by each panelist according to a replicated balanced complete block design (16 total samples tasted per session). Panelists were instructed to expectorate samples after tasting and cleanse their palates during a 2-minute enforced rest between samples using provided unsalted crackers and deionized water.

Degassing of HTST Milks

Following collection of data for light-exposed HTST and DSI-UP skim milks from analyses described above, a follow-up investigation of the role of DO was performed to assess if initial DO concentration was a significant determinant of LOF development. HTST milk was processed as previously described using a MicroThermics EHVH pasteurization unit (MicroThermics, Raleigh, NC) with a 2-stage homogenizer (GEA Niro Soavi, Parma, Italy), and was subsequently bottled in translucent, half-gallon, high-density polyethylene milk jugs (Upstate Niagara Cooperative Inc., Buffalo, NY) under an Aseptilab™ ultra-clean fill hood (MicroThermics, Raleigh, NC). Additionally, a second lot of HTST milk was prepared following the same processes, but was degassed using a vacuum chamber at 380mm Hg following the preheat step. HTST milks, both degassed (HTST-DG) and not (control), were stored for 48 h at 4°C and then subjected to light exposure as previously described for HTST milks. Milks were evaluated by trained panel and measured for DO and riboflavin at each experimental timepoint, as previously described. This process was repeated in triplicate.

Volatile Flavor Analysis

General volatiles from the milks were extracted via headspace solid phase microextraction (SPME). Five ml of milk were added to 20 ml amber vials, in triplicate, for each light exposure treatment. Next, 20 µl internal standard (deuterated hexanal in methanol, 507 ppm; Sigma Aldrich, Milwaukee, Wisconsin, U.S.A.) were added to each vial. Samples were placed on a CTC Analytics CombiPal Autosampler (Zwingen, Switzerland) and were subsequently analyzed on an Agilent 7820 gas chromatograph coupled with a 5975 mass spectrometer (GC-MS; Agilent, Santa Clara, CA, U.S.A.) with a ZB-5ms column (30m length x 0.25mm inner diameter x 0.25µm film thickness; Phenomenex, Torrance, CA). Initial analysis was conducted with a standard method scanning from 35-340 m/z to identify compounds of interest. Selected compounds were quantified using relative abundance in SIM mode.

Select sulfur compounds were also evaluated for each milk. Five ml of milk were added to 20 ml amber vials and 40 µL of internal standard (ethyl methyl sulfide in methanol at 1.65 mg/kg; Sigma Aldrich, St. Louis, MO, USA) was added to each vial. Volatiles were collected via SPME and subsequently analyzed using an Agilent 7890B gas chromatograph, coupled with an Agilent

7000C triple quadrupole mass spectrometer (GC-MS/MS) equipped with an Agilent sulfur selective flame photometric detector (FPD) with a ZB-5ms column. Sample introduction was conducted via a CTC Analytics CombiPal Autosampler as previously described (Jo et al., 2018). Analytical and operating conditions, including multiple reaction monitoring (MRM) transition of sulfur compounds and quantification of relative abundances were conducted as described by Jo et al. (2018).

Threshold Testing

Determination of the best estimate sensory threshold (BET) for LOF among the general population of fluid milk consumers was determined using a 7-series ascending forced choice methodology where the stimulus sample corresponded with light-exposed samples of increasing exposure time (1.5x step factor) and the other two samples were non-exposed controls from the same process run (ASTM, 2019). For each replicate of the experiment, 40 consumers were recruited from a database of over 10,000 consumers maintained by the North Carolina State University Sensory Service Center. In order to participate, panelists were required to report that they were regular consumers of fluid milk, and that they were regular purchasers/consumers of skim milk. Upon arrival, participants evaluated two sets (HTST and DSI-UP milk sets) of 7-series ascending triangle tests (threshold tests). Approximately 20ml aliquots of milk were served in lidded 60ml souffle cups coded with random three-digit blinding codes. Each series of each test was presented in a randomized order, balanced for signal position across all panelists. Tests (HTST or DSI-UP) were presented in a balanced randomized order for each panelist and a 5-minute enforced rest time was used between tests. Panelists were instructed to taste samples from left to right and pick the “odd” sample out of each row of samples by circling the corresponding blinding code on a paper ballot. A 30 s rest was then enforced after each series and panelists were instructed to cleanse their palates with provided unsalted crackers and deionized water before continuing to the next series. Threshold testing was conducted on milks at approximately 4°C.

Data Analysis

Analysis of variance (ANOVA) with Fisher's LSD post-hoc test for means separation was applied to data from the trained sensory and volatile compound analysis relative abundances to understand differences among the light exposure treatments. Linear regression was applied to dissolved oxygen and riboflavin degradation data and visualizations were constructed using the "ggplot2" package in R (Version 3.6.2) (Wickham, 2016). All statistical analyses were conducted at 95% confidence using XLSTAT software (Addinsoft, Paris, France).

The BET for LOF was determined for HTST and DSI-UP milks using the methods described by the American Society for Testing and Materials procedure E679-19 (ASTM, 2019). First, individual thresholds were determined by calculating the geometric mean of the highest concentration missed and the next concentration. In the case of a participant correctly identifying the odd sample for all rows, or incorrectly identifying the odd sample in the highest concentration row, the hypothetical next concentration above/below according to the 1.5x step factor was used to calculate the geometric mean for that individual. The group BET for LOF in both HTST and DSI-UP treatments was subsequently calculated by taking the geometric mean of all individual BETs for each test. Additionally, the standard deviation (SD) was calculated for each treatment from the $\log_{10}$ individual BETs to better understand variability in signal detection for LOF among untrained fluid milk consumers.

RESULTS AND DISCUSSION

Analysis of Riboflavin and Dissolved Oxygen

Photosensitized riboflavin in the presence of oxygen is well-recognized as the source of singlet oxygen oxidation of lipids and amino acids in fluid milk (Min and Boff, 2002). As such, measurement of riboflavin degradation and DO concentration is indicative of oxidation rate. Changes in riboflavin and DO concentrations over the course of experimental light exposure times for HTST and DSI-UP milks are depicted in Figures 2 and 3, respectively. Rate of riboflavin degradation was significantly greater ($p<0.05$) in HTST milk than DSI-UP milk. Likewise, the rate of decrease for DO was greater ($p<0.05$) in HTST milk than DSI-UP milk. Based on these findings, HTST milk appears to be more susceptible to light oxidation processes than DSI-UP milk. Initial concentrations of DO were also significantly greater in HTST milks, suggesting that rate of riboflavin degradation and DO decrease may be a function of initial DO concentration.

Trained Panel Sensory Profiling

Trained panel profiling of light-exposed milks was conducted at each experimental timepoint (Table 1). For HTST milk, the unexposed control was characterized by moderate intensities of sweet aromatic ($\bar{x}$ =2.3, SD=0.6) and cooked flavors ($\bar{x}$ =3.2, SD=0.2), and was devoid of cardboard or mushroom flavors (no LOF). LOF development in HTST skim milk was characterized by a decrease ($p<0.05$) (compared to unexposed control) in sweet aromatic flavor ($\bar{x}$ =1.3 , SD=0.3) and an increase in cardboard flavor ($\bar{x}$ =1.6 , SD=0.3) signal by 3.5 h, although a non-significant ($p>0.05$) signal for cardboard flavor was noted by the trained panel at 2.4 h. Significant ($p<0.05$) development of mushroom flavor was noted by 5.3 h ($\bar{x}$ =1.3 , SD=0.5). Observations by Whited et al. (2002) similarly observed noticeable flavor differences in skim and reduced fat milks after about 2 h of 2000 lx light exposure, which increased linearly in intensity with increased exposure time. In the present study, the onset of LOF in HTST milk was characterized by decreased sweet aromatic flavor intensity, and the development of cardboard and mushroom flavors. Following these timepoints, mushroom and cardboard flavor intensities continued to increase in intensity. Linear increases of LOF in fluid milk have also been reported by Chapman et al. (2002), although evaluations of light-exposed 1% milk by Brotherson et al. (2016) suggested that the presence of butterscotch flavor (a LOF attribute in fat-containing milks) may not follow a linear pattern. Over the 27 h total light exposure time used for HTST milk in the present study, no significant changes in cooked milk flavor, sweet taste, or astringency were noted ($p>0.05$). These results closely matched findings from Chang and Dando (2018), who reported sensory changes in skim milk following light exposure were primarily associated with light-exposed flavors (old oil, plastic, cardboard), not attributes inherent to unexposed skim milk.

DSI-UP milk was characterized by distinct sulfur/eggy flavors ($\bar{x} = 1.9$, SD=0.1 at 0 h exposure), which were not present in HTST milk, as well as higher cooked/milk flavors ($\bar{x} =3.9$, SD=0.1 at 0 h exposure) compared to HTST milk, consistent with previous studies (Lee et al., 2017; Jo et al., 2018). In addition, unexposed DSI-UP milk was devoid of any light-induced carboard or mushroom flavors. DSI-UP milk was largely similar to HTST milk in terms of flavor profile evolution over light exposure time, although the associated time domains were considerably greater in DSI-UP milk. Trained panelists identified LOF in DSI-UP milk after 36 h light exposure, with LOF being characterized by a decrease ($p<0.05$) in eggy/sulfur flavor ($\bar{x} =0.7$, SD=0.3), as well as an increase ($p<0.05$) in cardboard flavor ($\bar{x} =0.9$, SD=0.2). Previous studies have indicated that the decrease of eggy/sulfur flavors in UP/UHT milks is associated with the oxidation of sulfhydryl groups on associated compounds, indicating that these flavor compounds may play a direct antioxidant role, thus slowing LOF development (Adhikari and Singhal, 1992; Wadsworth and Bassette, 1985). Mushroom flavor development was not as pronounced in DSI-UP milk compared to HTST, even at extended exposure times, although a higher ($p<0.05$) signal ($\bar{x}$ =1.1, SD=0.1) (compared to unexposed control) was noted following 121.5 h light exposure. In general, previous sensory analysis studies of LOF in UP/UHT milks are rare, and, to our knowledge, none have described the specific flavor attributes relevant to LOF. However, the identification of LOF development in UP milk following 36 h light exposure is roughly in line with triangle test results of untrained panelists reported by Johnson et al. (2015), who noted differences ($p<0.05$) compared to a light-blocked control between 1d and 3d light exposure (~2186 lx) in 2% UP milk.

Volatile Flavor Analysis

HTST and DSI-UP milks were distinct in initial volatile compound profile, as expected (Jo et al., 2018). Thermal processing of skim milk via DSI-UP was characterized by higher ($p<0.05$) relative abundances, compared to HTST, for volatile sulfur compounds such as hydrogen sulfide, carbon disulfide, and dimethyl disulfide (Table 2). These findings were consistent with those reported by Jo et al., (2018), who identified such sulfur compounds as the source of potent sulfur/eggy flavors found in DSI-UP milk. Following 24 h light exposure, concentration of hydrogen sulfide had decreased ($p<0.05$) in DSI-UP milk, compared to 0 h light exposure measurements, before exhibiting an increasing trend thereafter. Similarly, dimethyl disulfide concentration was lower ($p<0.05$) at 54 h light exposure, compared to 0 h exposure, before increasing again. These trends suggest that sulfur compounds resulting from DSI-UP thermal processing likely imparted a protective antioxidant effect. While the antioxidant effects of such compounds has been addressed in previous fluid milk studies, those same compounds have been identified as secondary products of light-induced reactions and components of general LOF in other studies (Jung et al., 1998; Mestdagh et al., 2005). Due to their dichotomous role in the light oxidation process, changes in the concentration of such compounds may be a measurable indication of light-induced oxidation and terminal development of LOF in fluid milk and presents an ample area for future study.

The antioxidant effect of sulfur compounds inherent to UP or UHT milk thermal processing is a function of increased free sulfhydryl groups. Sulfhydryl group formation can be primarily linked to β-Lactoglobulin in serum protein, which is unfolded during heating (Liu et al., 2007). In the presence of UV light and singlet oxygen, oxidation reactions readily occur within fluid milk, destroying methionine and unsaturated fatty acids and resulting in the formation of oxidation-related volatiles (Min and Boff, 2002). When present in the fluid milk system, the sulfhydryl groups liberated in high heat treatments act as an intermediary, quenching free radicals and slowing the development of oxidation flavors. Salano-Lopez et al. (2005) noted that free sulfhydryl groups in UP milk were particularly reactive reducing agents and provided stability to ascorbic acid- another antioxidant natural found in milk- over the first 30 days of storage. Similarly, it is likely that the formation of sulfhydryl groups following DSI-UP thermal treatment resulted in decreased oxidation rate in the present study and delayed onset of LOF, as suggested by trained panel findings.

Other sulfur compounds commonly associated with LOF in HTST milk, such as dimethyl sulfide and dimethyl trisulfide, exhibited generally increasing trends. Dimethyl sulfide was higher ($p<0.05$) in concentration by 36 h light exposure, and dimethyl trisulfide was higher ($p<0.05$) in concentration by 54 h. While clear trends in relative abundances of sulfur compounds were observed, many of these changes were non-linear at extended light exposure times (54, 81, 121.5 h), further indicating that certain sulfur compounds play a significant role in secondary and tertiary light oxidation reactions (Al-Attabi et al., 2008; Jo et al., 2018).

Aldehydes and ketones commonly associated with oxidized flavor were also investigated (Brotherson et al., 2016; Jo et al., 2018). Initial (unexposed) concentrations of hexanal, heptanal, octanal, and nonanal were not different ($p>0.05$) in HTST and DSI milks. However, initial concentrations of methional, decanal, benzaldehyde, and 1-octen-3-one were greater in DSI-UP milk, consistent with Jo et al. (2018) that identified the presence of sulfur compounds, methyl ketones, and Maillard reaction products as key differentiators between DSI-UP and HTST milk. Over the course of light exposure, both HTST and DSI-UP milks exhibited increases ($p<0.05$) in aldehydes and ketones. In HTST milk, formation of cardboard and mushroom flavors determined by the trained sensory panel were correlated ($p<0.05$) with increases in methional ($r_{cardboard}=0.77$, $r_{mushroom}=0.78$), hexanal ($r_{cardboard}=0.82$, $r_{mushroom}=0.76$), heptanal ($r_{cardboard}=0.86$, $r_{mushroom}=0.82$), nonanal ($r_{cardboard}=0.82$, $r_{mushroom}=0.77$), and decanal ($r_{cardboard}=0.83$, $r_{mushroom}=0.84$). For DSI-UP milk, cardboard flavor was significantly correlated ($p<0.05$) with heptanal ($r=0.75$), nonanal ($r=0.86$), 2-nonanone ($r=0.74$), 2-heptanone ($r=0.76$), 1-octen-3-one ($r=0.81$), dimethyl sulfide ($r=0.74$), and dimethyl trisulfide ($r=0.92$), whereas mushroom flavor was significantly correlated ($p<0.05$) with increased 2-heptanone ($r=0.92$), 2-nonanone ($r=0.81$), 1-octen-3-one ($r=0.81$), and dimethyl trisulfide ($r=0.88$) concentrations. These findings are consistent with Whitson et al. (2010), who reported cardboard flavor of whey protein was associated with pentanal, heptanal, and nonanal in the presence of 1-octen-3-one and dimethyl trisulfide. Furthermore, identification of 1-octen-3-one as a primary source for mushroom flavor in UP milk is corroborated by gas chromatography-olfactometry findings reported by Jo et al. (2018). As 1-octen-3-one is formed by the thermal degradation of linoleic acid (Lin and Blank, 2003), it is unsurprising that this characteristically mushroom-like volatile compound is present at initial higher abundance in DSI-UP milk compared to HTST and may play a more direct role in LOF in UP milk.

Threshold Determination

Estimation of the BET for retronasal detection of LOF in both HTST and DSI-UP skim milk was determined following data collection with untrained self-reported skim milk consumers (Figure 4). For HTST skim milk, the population BET was 15.2 h (SD=2.3 h), whereas the BET for DSI-UP milk was higher ($p<0.05$) at 61.0 h (SD=2.0 h). For the HTST treatment, results generally agreed with Chang and Dando (2018), who reported a LOF development threshold of approximately 9 h in HTST skim milk under LED light when assessed by untrained consumers. However, light exposure times associated with the determined BET values from this study are greater than the estimated 2 h BET for LOF in HTST reduced fat milk exposed to 2,000 lx (± 5%) reported by Chapman et al. (2002). Similarly, Walsh et al. (2015) reported HTST 2% fat milks exposed to 375 lx fluorescent light for 8 h received significantly lower liking scores than unexposed controls, suggesting that consumers detected LOF in the light exposed milks. It should be noted that the present study differed from the aforementioned studies concerning fat content, light type, and threshold determination method; however, the magnitude and direction of observed differences compared to our study with skim milk implies that fat content may play a significant role in flavor degradation from light exposure. Whited et al. (2002) observed reduced fat and whole milks exhibited more rapid LOF development compared to skim milk, seemingly contradicting findings within the same study that showed skim milk to be more susceptible to riboflavin and vitamin degradation. Further investigation into the interaction of fat content, light exposure, and other variables is warranted to more thoroughly explore and explain these tendencies in fluid milk.

The greater ($p<0.05$) BET observed for DSI-UP milk compared to HTST milk, along with consistent trained panel and riboflavin degradation results, demonstrates that thermal processing at the extreme temperature of DSI-UP impacts LOF. Sulfur-containing compounds associated with UP treatment of fluid milk have been directly linked to the unique cooked and sulfur/eggy flavors of UP milk (Jo et al., 2019). As these flavors/aromas tended to be more intense than typical light oxidation flavors (mushroom, cardboard) at early light exposure times, it is likely that the extended threshold for LOF observed for DSI-UP skim milk is a function of masking effects. Sensory masking is a phenomenon where intensity of a sensory signal for a particular component is lessened by the presence of a secondary component. Lawless (2000) noted that sensory masking effects often result in mutual suppression of the components within a system. Within HTST milks, development of LOF seemingly coincided with decreased perception of typical sweet aromatic flavor, when evaluated by trained sensory panelists. Similarly, this inverse relationship was mirrored by LOF and eggy/sulfur flavors from DSI-UP processing. Previous studies with ultrapasteurized milk have noted that American milk consumers tend to dislike UP/UHT milks because of the presence of sulfurous flavors, and incongruence with HTST milk flavors with which consumers are more familiar (Chapman and Boor, 2001; Lee et al., 2017). Effects of flavor suppression have been found to be more pronounced when incongruent flavors are present together (Frank et al., 1991). In this way, the presence of eggy/sulfur flavors may be responsible for the delayed onset of LOF detection in DSI-UP milks as they mask typical off-flavors at early stages of light exposure.

Degassing of HTST Milk

Inclusion of a degassing step resulted in a lower ($p<0.05$) initial DO concentration for the HTST-DG treatment, compared to the HTST control. However, it should be noted that the decrease in DO achieved through the inclusion of a degassing step was not equal ($p<0.05$) to the level of DO measured in DSI-UP milk. For the DSI-UP treatment, the addition of water and high heat to the system necessitated a greater vacuum (1040mm Hg) for flash vacuum cooling than was possible or appropriate for the HTST treatment (380mm Hg). A greater vacuum reduces vapor pressure, leading to more efficient removal of dissolved gases within the milk; similarly, higher temperatures reduce oxygen solubility, aiding in the vacuum cooling/degassing process (Carlsson and Jönsson, 2012). The rate of DO decrease in fluid milk is widely considered an indicator of oxidation (Potts et al., 2017). To our knowledge, no previous works have assessed the effect of different dissolved oxygen concentrations in HTST milk. However, previous studies on UHT milk have suggested that milk deaeration had no impact on riboflavin stability during storage (Oamen et al., 1989). Comparison of HTST-DG versus HTST milk was consistent with such findings, as the rate of riboflavin degradation was unchanged when post-processing DO was decreased (Figure 2). Additionally, rate of DO consumption between HTST and HTST-DG milk treatments was not different ($p>0.05$). Trained panel sensory evaluations mirrored riboflavin/DO monitoring, as no significant difference ($p>0.05$) in the formation of LOF was found between HTST and HTST-DG milk treatments. Overall, these findings suggest that the lowering of DO alone does not significantly affect the development of LOF in fluid milk at the levels achievable with HTST processing. However, degassing steps should not be considered unserviceable, as higher initial DO concentrations in UHT fluid milk are associated with greater vitamin losses during extended storage (Oamen et al., 1989).

CONCLUSIONS

Oxidation following exposure to light has been a long-known source of off-flavor for fluid milk and dairy products. Mitigation of off-flavor development due to light exposure has often been addressed via antioxidant addition, or the use of light-shielding materials; however, there has been limited investigation on the role thermal processing plays in the development of LOF in fluid milk. The present study found that DSI-UP milk is associated with a significantly greater ($p<0.05$) time until the threshold for LOF is reached (61.0 h light exposure), compared to HTST milk (15.2 h light exposure). The observed difference in time until LOF onset by consumers was corroborated by a trained sensory panel, who detected LOF in DSI-UP milks after 36 h light exposure, compared to only 3.5 h in HTST milk. Evidence suggests that the delayed threshold for DSI-UP milk can be attributed to higher relative abundance of volatile sulfur compounds, which result in sensory masking of early LOF development and possibly impart an antioxidant effect as well. Overall, results from this study demonstrate the advantages of ultrapasteurized milk compared to HTST milks in terms of delayed LOF onset, and provide valuable information that may influence future packaging, storage, and retail presentation for such products.

ACKNOWLEDGEMENTS

Funding was provided in part by the National Dairy Council (Rosemont, IL). The use of trade names does not imply endorsement or lack of endorsement of those not mentioned.

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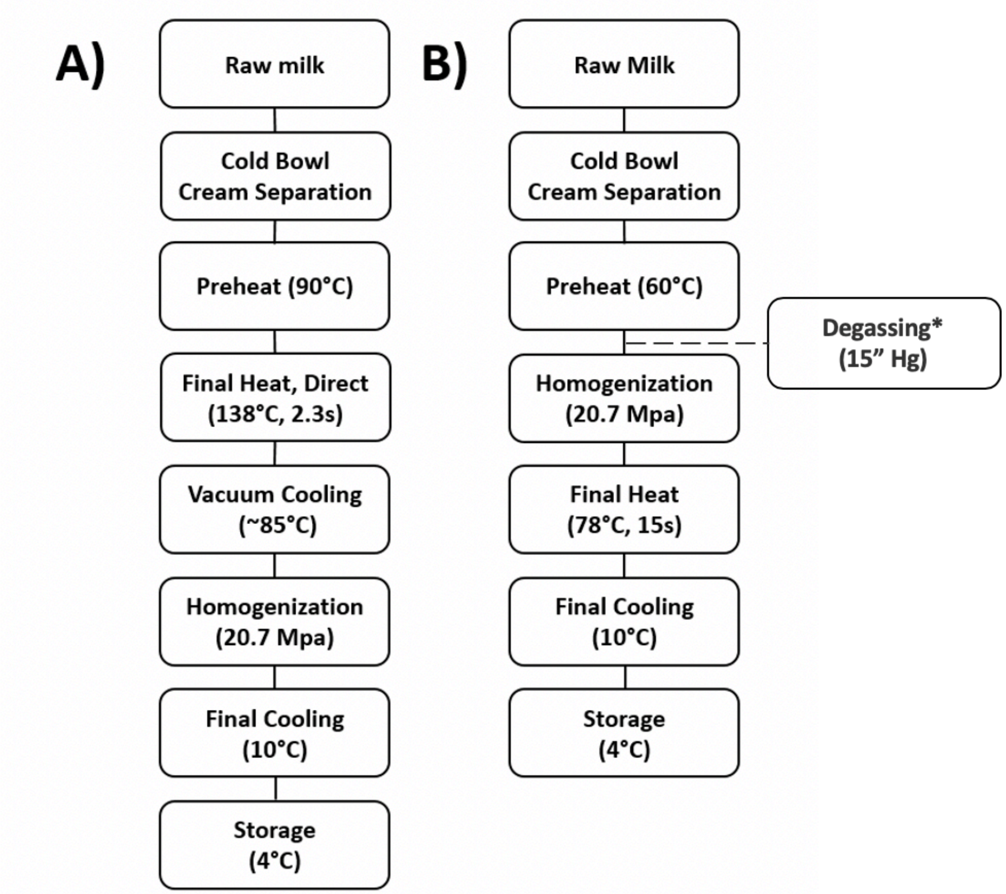

Figure 1. Process flow diagram for A) direct steam injection ultrapasteurization (DSI-UP) and B) high temperature short time (HTST) skim milk treatments

*Degassing of HTST-processed milks was only conducted for HTST vs Degassed HTST (HTST-DG) treatment comparisons, not as a part of HTST vs. DSI-UP treatment comparisons

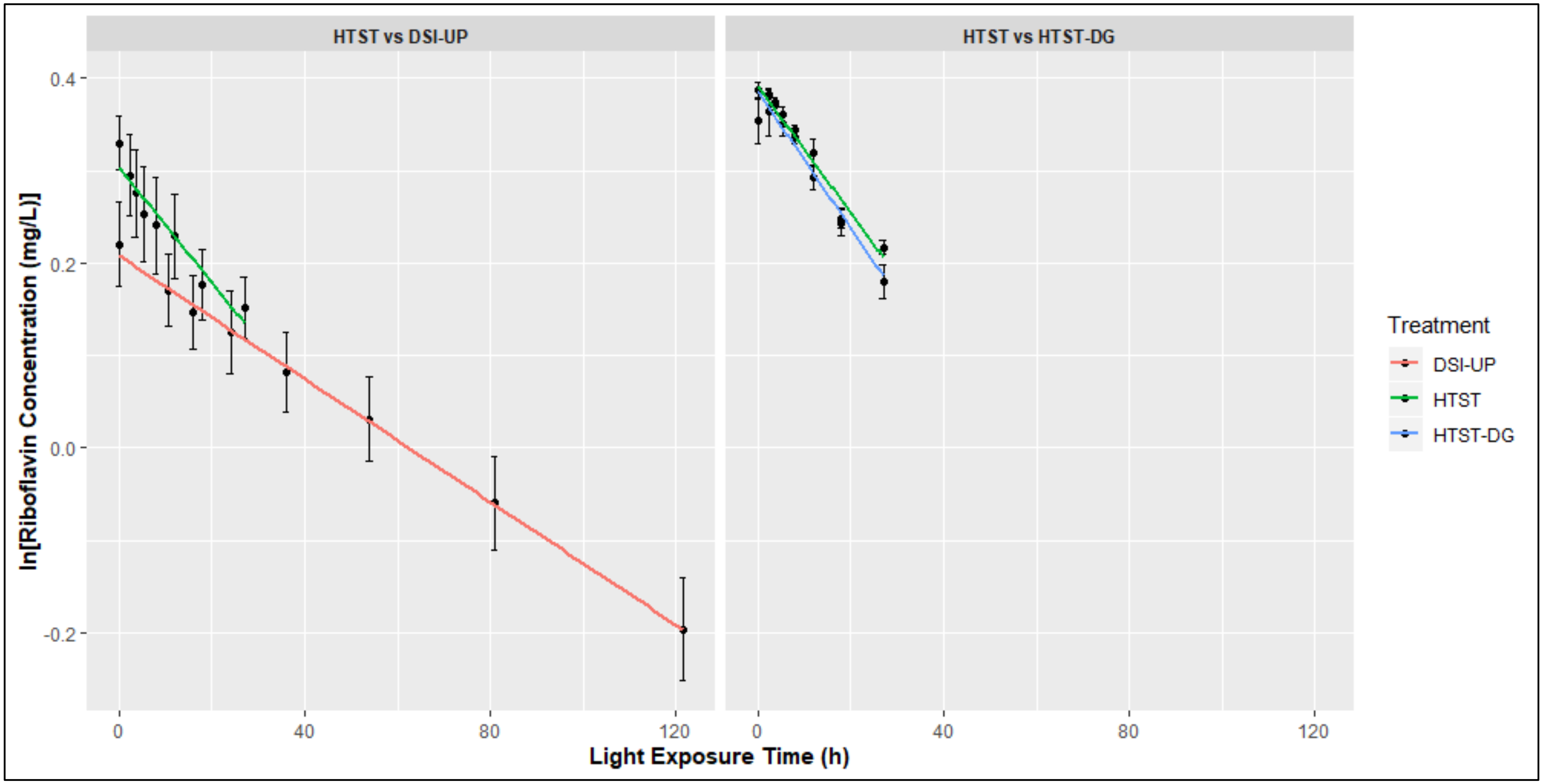

Figure 2. Riboflavin degradation over light exposure time for high temperature short time (HTST) vs. direct steam injection ultrapasteurization (DSI-UP) and HTST vs. degassed HTST (HTST-DG) skim milk treatments.

Data points represent mean riboflavin concentration measurements and bars represent standard error of the mean over three replicates at each experimental light exposure duration for each treatment.

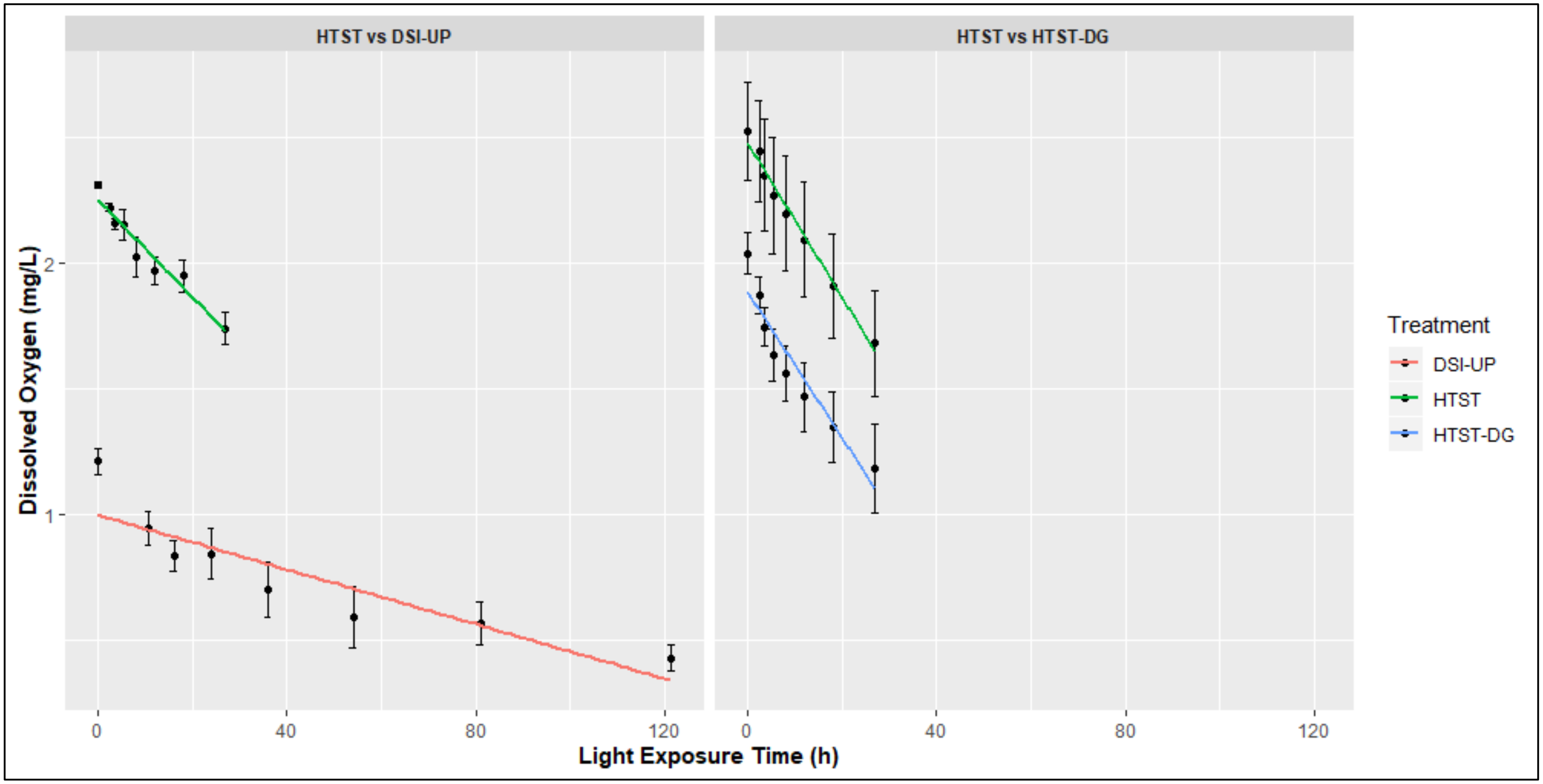

Figure 3. Dissolved oxygen measurements over light exposure time for high temperature short time (HTST) vs. direct steam injection ultrapasteurization (DSI-UP) and HTST vs. degassed HTST (HTST-DG) skim milk treatments.

Data points represent mean dissolved oxygen measurements and bars represent standard error of the mean over three replicates at each experimental light exposure duration for each treatment.

Table 1. Descriptive sensory profiles of control and light-exposed high temperature short time (HTST) and direct steam injection ultrapasteurization (DSI-UP) skim milk treatments determined by trained panel (n=8)

HTST					DSI-UP					
Light Exposure Duration (h)	Sweet Aromatic	Cooked	Cardboard	Mushroom	Light Exposure Duration (h)	Sweet Aromatic	Cooked	Cardboard	Mushroom	Eggy/ Sulfur
0 (Control)	2.3	3.2	ND	ND	0 (Control)	1.3	4.0	ND	ND	1.9
2.4	1.8	3.0	0.7	ND	10.7	1.3	4.0	ND	ND	1.9
3.6	1.3	3.0	1.6	0.7	16	1.3	3.9	ND	ND	1.7
5.3	0.8	3.0	2.2	1.3	24	1.3	3.9	ND	ND	1.5
8	0.8	3.0	2.3	1.3	36	1.0	3.7	0.8	ND	0.7
12	0.5	2.9	2.7	1.9	54	1.0	3.7	0.7	ND	0.7
18	0.6	2.9	2.8	2.0	81	ND	3.5	1.6	ND	ND
27	ND	2.9	3.3	2.2	121.5	ND	3.4	1.8	1.1	ND
*LSD**	*0.74*	*0.17*	*1.05*	*0.95*	---	*0.73*	*0.39*	*0.56*	*0.11*	*0.85*

ND=Not detected, or insignificant signal (<0.5 on 0-15 point universal intensity scale)

*LSD= Fisher’s Least Significant Difference (α=0.05)

**Not shown- sweet taste and astringency were also measured by trained sensory panel; however, no differences in intensity related to either attribute were noted. Eggy/sulfur was not noted in the HTST treatment milks.

Table 2. Relative abundances (ppb) of selected compounds in control and light-exposed high temperature short time (HTST) and direct steam injection ultrapasteurization (DSI-UP) skim milk treatments

	HTST								**DSI-UP**								
Compound	**0h**	**2.4h**	**3.6h**	**5.3h**	**8h**	**12h**	**18h**	**27h**	**0h**	**10.7h**	**16h**	**24h**	**36h**	**54h**	**81h**	**121.5h**	**LSD**
Methional	32.37	28.49	46.44	33.57	46.84	44.25	57.19	71.98	60.43	87.67	68.09	36.17	49.53	46.58	65.18	55.88	25.95
Hexanal	51.54	180.10	154.77	206.46	301.90	275.73	186.73	277.90	18.65	34.97	56.73	87.63	117.60	303.20	179.84	171.86	83.25
Heptanal	2.79	6.38	5.57	7.15	9.50	9.34	7.53	14.20	2.02	4.00	5.24	9.13	15.07	45.57	63.32	25.83	17.92
Octanal	1.71	2.22	2.15	2.48	3.20	2.55	1.65	3.32	0.97	2.42	3.59	4.15	5.53	13.93	25.27	6.31	6.22
Nonanal	3.63	7.02	6.13	7.07	8.99	8.63	6.70	10.09	3.00	5.12	6.34	8.88	15.47	34.56	58.44	32.77	19.60
Decanal	1.40	1.83	1.57	1.98	2.69	2.36	3.62	4.57	5.23	6.42	6.12	8.21	4.43	3.35	4.33	3.87	2.64
Benzaldehyde	13.14	11.71	10.82	9.56	8.50	8.47	7.52	6.86	24.71	29.66	31.06	34.96	34.27	36.29	52.16	28.72	11.14
2-Heptanone	16.14	12.69	14.08	13.07	12.95	12.55	12.82	14.79	8.65	12.72	9.09	10.72	10.69	12.23	13.36	30.15	6.62
2-Nonanone	18.93	14.61	16.24	15.70	14.25	14.81	15.01	15.85	12.72	14.70	14.32	14.78	13.92	14.71	15.44	16.65	4.96
1-octen-3-one	32.56	56.29	49.85	47.64	45.71	51.13	80.73	69.43	116.36	125.01	126.85	154.04	137.88	121.04	190.67	179.57	85.52
Hydrogen sulfide	4.38	55.56	26.24	33.98	30.45	56.44	64.04	14.44	232.82	228.81	173.27	100.46	146.94	189.20	154.74	235.97	61.68
Carbon disulfide	50.61	67.67	42.74	52.30	61.18	50.78	43.39	48.57	131.45	146.01	169.60	152.69	194.63	188.21	207.17	125.48	50.85
Dimethyl sulfide	53.53	56.31	57.19	86.47	79.52	64.64	95.75	38.98	10.18	18.99	25.34	51.06	80.70	144.13	65.99	161.16	50.06
Dimethyl disulfide	0.34	0.23	0.33	0.34	0.42	0.32	0.50	1.19	1.54	0.47	1.39	0.93	0.89	0.74	1.84	1.36	0.71
Dimethyl trisulfide	0.30	0.44	0.46	0.36	0.31	0.42	0.31	1.69	0.94	1.19	3.36	2.48	2.19	3.84	8.76	8.50	2.10

* Means within a row that differ by the least significant difference (LSD) are different within heat treatment and hours of light exposure ($p < 0.05$).

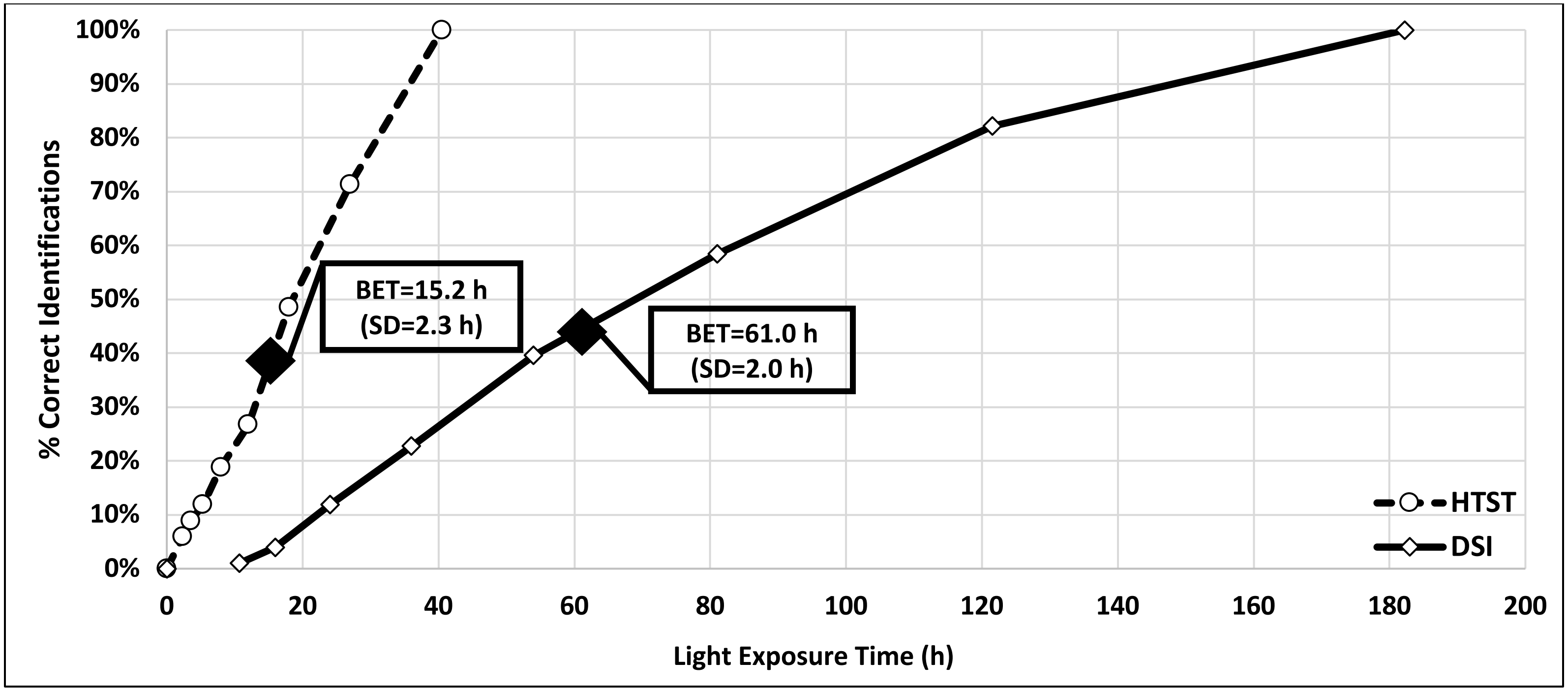

Figure 4. Consumer best estimate threshold (BET) determination from three experimental replications (n=101 total evaluations) of light oxidized flavor (LOF) for high temperature short time (HTST) and direct steam injection ultrapasteurization (DSI-UP) skim milks.

SD=standard deviation

CHAPTER 4: The Influence of Automatic Associations on Preference for Fluid Milk Types

The Influence of Automatic Associations on Preference for Fluid Milk Types

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The content of this chapter was submitted for publication in:

Journal of Dairy Science (in press)

ABSTRACT

In recent decades, organic milk has been an exception to the trend of decreased fluid milk consumption in the US. However, the reasons behind consumer preference for organic milk over conventional and other milk types are ill-defined. The objective of this study was to use an Implicit Association Test (IAT) and primed/unprimed preference testing to determine if fluid milk consumer preferences for milk types are influenced by implicit biases and, if so, to define these biases within the context of the consumer sensory experience. Self-reported fluid milk consumers (n=473) participated in online IAT exercises where pairwise comparisons of milk types (conventional, organic, local, pasture-raised) were measured on both positive and negative dimensions related to cow-welfare, sustainability, health, trust, safety, and quality. Latency times from IAT responses were subsequently transformed into standardized D scores to categorize bias effect sizes. Additionally, fluid milk consumers (n=174) participated in preference tests where commercial milks representing different milk types were compared in presentations where milk type was shown (primed) and hidden (unprimed). Following preference tests, consumers were asked to explain their preferred sample using check-all-that-apply tasks. Analysis of IAT results showed that conventional milk was more associated ($p<0.05$) with negative dimension descriptors compared to organic, local, and pasture-raised milks. Additionally, a positive bias in favor of organic milk was expressed when compared to pasture-raised milk offerings; however, no differences ($p>0.05$) were found in pairwise comparisons of other non-conventional milk type pairings, suggesting that consumers may conflate these designations. Blinded preference testing showed that milk preferences were largely dictated by flavor, with consumers differentiating milk types based upon flavors related to packaging, pasteurization method, and feeding systems. In primed evaluations, consumers generally expressed preferences that aligned with their explicit

beliefs, and flavor considerations appeared to be a secondary differentiator of preference. Based on these results, conventional milk is associated with negative implicit beliefs related to production and product quality more often than other milk types, which were reflected in IAT evaluations and primed preference tests. However, blinded tastings suggest that conventional milk was preferred, or competitive with, other milk types based on flavor alone. Findings from this study suggest consumer differentiation and preference of milks is significantly impacted by perceptions/beliefs related to milk type. In particular, conventional milk was associated with relatively few unique belief descriptions, indicating strict utilitarian consumer categorization. Organic and local milks were comparatively associated with greater care for nutrition, sustainability, animal welfare, and local farm support. These results demonstrate a need for greater education related to conventional milk offerings to dispel factors influencing negative implicit bias. Furthermore, improving product narrative via label information and alignment with locality/regionality were identified as possible opportunities for improving consumer sentiments related to conventional milk. A more thorough understanding of these attributes may reinforce stated beliefs more effectively and stave off consumer losses to plant-based alternatives that fulfill similar beliefs.

INTERPRETIVE SUMMARY

Consumer preferences are dictated by both explicit and implicit motivations. Assessing both explicit and implicit consumer sentiments as they relate to milk type may provide guidance for fluid milk producers and farmers, and may guide more effective marketing and education campaigns for the milk industry. The present study utilized implicit association tests in conjunction with consumer preference tests to better understand the patterns and belief systems that dictate consumer choice of commercial fluid milks.

Keywords: milk, organic, implicit, bias, local, consumers

INTRODUCTION

Consumption of fluid milk has steadily declined the last several decades, coinciding with increased consumption of plant-based alternatives (USDA ERS, 2019; Haas et al., 2019). While this trend is problematic for milk producers, fluid milk is still widely considered a staple food item in many American households (McCarthy et al., 2017). Furthermore, commercial fluid milk offerings have grown considerably over the last few decades to meet consumer needs. For example, growing consumer concern about lactose intolerance has been widely addressed through the production of lactose-free milks via lactase enzyme or ultrafiltration in such a way that multiple consumer sentiments may be addressed through available commercial milk options (Rizzo et al., 2020). Similarly, technological advances in packaging and increased thermal treatment have helped assuage consumer concerns related to shelf-life of fluid milk, although application of higher heat treatments alter the sensory profile of milk (Gandy et al., 2008; Lee et al., 2017).

While flavor preference is certainly a driver of ultimate acceptance, previous studies on milk and milk alternative consumers have indicated that the primary reasons for choosing plant alternatives include greater perceived animal welfare, greater environmental sustainability, and better nutrition (McCarthy et al., 2017; Haas et al., 2019). However, non-conventional fluid milk offerings such those designated as organic, locally-farmed, or pasture-raised may similarly comply to these consumer belief systems. In a survey of fluid milk consumers, those who exclusively consumed organic milk reported doing so largely because they perceived greater health and quality compared to conventional milk, and believed organic milk production was associated with greater support for local farms/farmers, higher animal welfare, and greater sustainability (Harwood and Drake, 2018). Furthermore, findings from Harwood and Drake

(2020) reported that milk consumers valued the increased shelf life of organic and pasture-raised milks that were ultrapasteurized, though consumers were largely unaware these qualities were associated with increased thermal processing treatment. Cardoso et al. (2016) additionally reported that pasture access, adherence to organic practices, and engagement with local farming were cited as essential parts of an “ideal” dairy farm in open-ended consumer surveys, reinforcing consumer attraction to such features. Quantification of these sentiments as they relate to commercial fluid milks has been extensively studied, however, these findings are often limited to willingness-to-pay (WTP) and price modelling studies (Wolf et al., 2011; Kühl et al., 2017). As such, a more detailed understanding of consumer perceptions related to different dairy systems is warranted in order to inform product marketing, education initiatives, and potential benefits/risks of transitioning farming practices.

Several methods exist for capturing consumer beliefs, preferences, and motivations; however, this plethora of tools can generally be condensed to either implicit or explicit measures. Explicit beliefs generally refer to conscious attitude or beliefs that are directly expressed by an individual upon introspection, whereas implicit beliefs are characterized as subconscious, underlying, unknown, or otherwise unable to be readily expressed (Friese et al., 2006). Explicit belief measures commonly used for food products include direct rating (hedonic, magnitude estimation, Likert, semantic differential, or agreement scales), choice procedures (preference tests), or free-response to capture conscious motivations and preferences (Moskowitz and Sidel, 1971; Lim, 2011). Although explicit measures are incredibly useful in modeling and defining preferences, consumers may hold subconscious implicit motivations that also guide their ultimate behavior (Friese et al., 2006). Implicit measures can be collected in a variety of ways, including physical responses or multinomial choice experiments (Ares et al., 2013; Harwood et al., 2019).

Furthermore, the joint application of explicit and implicit measurements may provide a more holistic understanding of consumer preferences and may help correct for incorrect assumptions or misleading conclusions. The present study proposes the application of this dual approach to understand consumer preferences as they relate to fluid milk farming systems from an explicit and implicit perspective, coupled with insights that address both sensory and conceptual motivations.

MATERIALS AND METHODS

Experimental Overview

Implicit beliefs, sensory preferences, and primed preferences associated with fluid milk types (conventional, organic, locally-farmed, pasture-raised) were assessed using a two-part study. In the first part of the study, an online implicit association test (IAT) was used to compare fluid milk type pairings to evaluate the presence or absence of implicit biases. IAT participants (n=473 total) were asked to complete three of a possible six IAT tests (representing the 6 possible milk type pairings). Participants who completed all three assigned IAT exercises were entered into a drawing to receive a gift certificate to a local store. In the second part of the study, fluid milk consumers (n=174 total) were recruited to participate in a series of six preference tests, representing the six possible milk type pairings. Preference test participants were randomly assigned to either a blinded, or primed (milk types disclosed) group in order to understand the effect of milk type on preferences. Preference test participants were compensated with a $10 gift certificate to a local store. All procedures were conducted in compliance with North Carolina State University Institutional Review Board regulations.

Implicit Association Test (IAT)

Six online IAT surveys, each representing a possible milk type pairing, were constructed using the iatgen web-based tool developed by Carpenter et al. (2019) and were hosted on the Qualtrics (Provo, UT) software platform. Each IAT survey consisted of two milk type targets, which were each represented by six images of commercial fluid milks corresponding to the given type, and two categories, representing positive and negative attributes related to milk production. Positive category attributes were the following: happy cows, sustainable, healthy, trustworthy, safe, high-quality. Negative category attributes were the following: unhappy cows, wasteful, unhealthy, misleading, unsafe, low-quality. Selection of category attributes was intended to capture multiple angles related to milk production (animal welfare, sustainability, health, quality, etc.), and terminology was influenced by previous studies related to fluid milk and milk production beliefs (Cardoso et al., 2016). An additional survey was composed to capture basic demographic and usage questions prior to participation in the IAT surveys, as well as to screen out unqualified participants. Participants who were at least 18 years of age and reported consumption of fluid milk at least “a few times per month” were considered for participation in the IAT tests. Following completion of demographic and usage questions, qualified participants were randomly assigned three of a possible six links, with each link corresponding to a different IAT.

Each IAT consisted of seven blocks of sorting tasks, as described by Carpenter et al. (2019), which were randomized and balanced to account for order effects (Nosek et al., 2005). An example overview for the conventional vs. organic milk type pairing IAT can be found in Table 1. For each test, participants were introduced to the stimuli (milk type images and positive/negative text attributes) prior to beginning the IAT exercise. For each block, respondents were asked to sort (using the ‘e’ key to sort left, or the ‘i’ key to sort right) stimuli to the left or right side of their screen to match the appropriate category, which was fixed to a given side. For example, the screen may show an image of an organic milk in the middle of the screen, with the term “Organic” appearing on the left side of the screen, and “Conventional” appearing on the right side of the screen. The panelist would then need to press the ‘e’ key to sort the image correctly to the “Organic” category. In the case of an incorrect answer, an error message immediately appeared, and panelists were only able to move on once the stimulus was correctly answered. Within the fourth and seventh blocks (scored combined blocks), latency time for each sorted stimulus was recorded. In the event of an incorrect answer, latency time was recorded once the stimulus was correctly sorted, with no penalty (Greenwald et al., 2003). For the purposes of replicating these methods, or adopting the approach for IAT survey design described in the present study, the reader is directed to review Carpenter et al. (2019)- a comprehensive overview on survey-based IAT design using the iatgen web tool.

Preference Testing

Self-reported fluid milk consumers (n=174 total) were recruited to participate in forced-choice preference tests (2-AFC) on commercial milks representing different fluid milk types. Milks chosen for the study represented conventional (Harris Teeter 2%; store brand, high temperature short time pasteurization, HDPE jug), organic (Harris Teeter Organic 2%; store brand, ultrapasteurized, paperboard carton), locally-farmed (Maple View Farms 2%; high temperature short time pasteurization, glass bottle), and pasture-raised (Organic Valley Grassmilk 2%; national brand, ultrapasteurized, paperboard carton) milk types. Upon arrival, panelists were randomly assigned to either a blinded (unprimed) cohort (n=87), or a primed cohort (n=87). Panelists then proceeded to perform six 2-AFC tests on each possible milk type pairing. Presentation of milk pairings was randomized and balanced for sample position across consumers, and evaluations were performed and collected on an iPad interface hosting Compusense Cloud software (Guelph, Ontario). Prior to serving, each container of commercial milk used in the study was screened by two researchers (each with >250 hrs trained panel experience with milk and dairy products) to ensure no undesirable/unacceptable alterations to flavor or quality dur to light oxidation, spoilage, etc. were present. Milks were served at approximately 4°C in 177 mL white Styrofoam cups labeled with 3-digit blinding codes. For blinded panelists, only the 3-digit blinding codes were displayed. Primed panelists similarly received samples with 3-digit blinding codes, but were also made aware of what the corresponding milk type was for each sample using text labels on the data collection interface. Text descriptions of milk type for primed consumers disclosed the milk type designation (conventional, organic, locally-farmed, pasture-raised), but made no mention of brands, logos, or other label features. Once sample pairs were received, panelists were instructed to taste both samples, left to right, before deciding which sample they preferred. Following evaluation of each milk pair, a 3 min rest time was enforced, during which time panelists were instructed to cleanse their palate with water and an unsalted cracker. Once finished with evaluation of all six milk preference tests, panelists were asked to identify what features typically influenced their purchase of fluid milk using a list of 22 features in a check-all-that-apply (CATA) format.

Data Analysis

Following data collection, raw latency times from IAT survey sorting tasks were subjected to cleaning and analysis using the iatgen web-based analysis tool (Carpenter et al., 2019). Latency times over 10,000 ms were scored as missing, as were observations from participants who had >10% trials under 300 ms (indicating a “click-thru” series of responses).

Cleaned data were used to determine D-scores for each respondent using the algorithm described by Greenwald et al. (2003). Lesser latency times (shorter response times) for targets (milk types) when combined with the positive descriptors category, compared to when those same targets were combined with negative descriptor category, were indicative of positive implicit biases. Additionally, the same was true for the other milk type being concurrently assessed, allowing for a relative measure of implicit bias for each milk type pairing.

In order to calculate D-scores, within-person difference scores were first calculated by determining the mean latency time of combined block practice results (blocks 3 and 6) and combined block scored results (blocks 4 and 7). Next, these mean values were each divided by their associated standard deviations (SD), and the two resulting values were averaged to create a D-score, similar in nature to calculation of Cohen's *d* for determining effect size. Individual D-scores were used to determine population statistics for each IAT (mean, SD, Cohen's *d*), and were also stratified based on magnitude to describe general strength of observed biases as non-existent, slight, moderate, or strong (Greenwald et al., 2003; Rooth, 2010). Analysis of variance (ANOVA) of D-scores with gender, age, presence of children, annual household income, ethnicity, and stated milk type preferences as independent variables was subsequently conducted to investigate demographic determinants of milk type bias. Milk type preferences were asked in a check-all-that-apply (CATA) format, with conventional, organic, locally-farmed, and pasture-raised milk types shown as available options. Results from this CATA question were subsequently condensed to define consumers as exclusive conventional milk consumers (only conventional chosen), exclusive non-conventional consumers (only non-conventional options chosen), or mixed option consumers (both conventional and non-conventional consumption habits).

Blinded and primed consumer preference test results were tabulated and subsequently analyzed using the binomial test (α=0.05) to determine if significant differences in preference existed between milk types. Additionally, relationships between milk type preferences (both blinded and primed) and factors influencing fluid milk purchase (CATA) were assessed using the multiple correspondence analysis (MCA; rows= 87 panelists for each test condition, columns=6 preference test results; 22 CATA features related to belief systems and purchase preference were included as supplemental variable columns) module within XLSTAT version 2020.1.2 (Addinsoft, Paris, France). Coordinates derived from MCA were subjected to agglomerative hierarchical clustering using a dissimilarity matrix with Euclidean distance, Ward's agglomeration method, and entropy-based truncation of segments in order to effectively communicate connections between preference test results and CATA-stated beliefs.

RESULTS AND DISCUSSION

Implicit Association Test (IAT)

Across the six IAT tests, approximately 8.4% of evaluations were dropped due to incompletion or excessive speed within exercises (Table 2). Within the final dataset, D-scores were calculated to assess whether an implicit bias existed between milk type pairings. It should be noted that a positive or negative D-score is indicative of a directional implicit bias in favor of the milk type associated with linked direction. For example, within the conventional/organic milk type pairing, conventional was designated as the positive (+) direction milk type and organic was designated as the negative (-) direction milk type. In this case, the negative D-score for this pairing indicates an implicit bias directionally in favor of the organic milk type designation. An overview of positive/negative direction designations within each milk type pairing can be found in Table 2.

Analysis of IAT D-scores showed conventional milk was consistently associated with negative milk production attributes and farm practices, compared to organic (D=-0.26, *p*=<0.001, *d*=-0.46), locally-farmed (D=-0.28, *p*=<0.001, *d*=-0.48), and pasture-raised (D=-0.23, *p*=<0.001, *d*=-0.38) milk types (Table 2). Evaluation of effect sizes and stratified individual results indicated that these biases may be interpreted as being moderate in nature, although strong biases in favor of non-conventional offerings were observed as the largest classification group for each test. Investigation of ANOVA main effects for each milk type pairing (results not shown) indicated that stated milk type preferences were a significant determinant ($p<0.05$) of biases in conventional vs. non-conventional milk type pairings, with respondents who reported strict conventional milk purchase habits expressing a lower ($p<0.05$) bias in favor of organic, locally-farmed, or pasture-raised, compared to respondents who reported strict non-conventional or mixed (conventional and non-conventional) milk type purchase habits. However, it should be noted that in each convential pairing, respondents with strict conventional milk purchase habits directionally expressed biases against conventional milk. These results are consistent with findings from Hill and Lynchehaun (2002), who suggested that purchase habits of conventional milk consumers was more habitual, compared to organic milk consumers who tend to exhibit more thoughtful and loyal purchase habits. Additionally, gender was reported as a significant determinant ($p<0.05$) of bias for the conventional/pasture-raised pairing, with female respondents reporting a greater ($p<0.05$) pro-pasture-raised bias compared to male respondents. This result can likely be attributed to the suggestion that pasture-based systems offer greater animal welfare, which has been shown to be a significantly more impactful issue for women than men (Vanhonacker et al., 2007).

Comparisons of non-conventional milk types yielded mixed results. A significant implicit bias, albeit "slight" in magnitude, was observed in favor of organic milk when compared to pasture-raised (D=0.10, *p*=0.001, *d*=0.22). However, no bias was found in organic/locally-farmed (D=-0.02, *p*=0.541, *d*=-0.04) or pasture-raised/locally-farmed (D=-0.03, *p*= 0.409, *d*=-0.06) pairings. Analysis of main effects from the ANOVA model for the organic/pasture-raised pairing indicated the presence of children in the household and respondent ethnicity were significant determinants of bias. Specifically, a greater ($p<0.05$) pro-organic bias was expressed for respondents who had children (compared to those who did not have children in their household), as well as for non-Caucasian respondents (compared to Caucasian respondents), consistent with trends reported in previous studies (Harwood and Drake, 2018). For the pasture-raised/locally-farmed pairing, income and ethnicity were identified as significant determinants ($p<0.05$) of bias. Specifically, greater ($p<0.05$) pro-locally-farmed bias was expressed by non-Caucasian respondents, as well as high-earners (>$80,000/year), compared to moderate earners ($40,000-$80,000/year), although there was no difference in bias ($p>0.05$) compared to low earners (<$40,000/year). Finally, for the organic/locally-farmed pairing, no differences were noted among demographic groups, suggesting widespread belief that the two designations are interchangeable, or share core features (Haas et al., 2013).

Preference Testing

Evaluation of preference test results from blinded and primed consumer cohorts demonstrated sensory preferences, as well as the effects of milk type labeling for commercial fluid milks (Table 3). Within blinded evaluations, organic milk was preferred ($p<0.05$) to conventional milk and locally-farmed milk was preferred ($p<0.05$) to pasture-raised milk. Within primed evaluations, no significant preferences ($p>0.05$) were noted for any milk type pairing.

Furthermore, chi-square tests on proportions from blinded and primed tests revealed no significant differences in preference due to priming. Differences in preference observed from blinded preference tests suggest that meaningful differences in sensory profiles exist in milks from different dairy systems; however, primed evaluations show that independent conclusions about these differences may be altered by the presence of milk type labeling.

Discussion

When viewed independently, differences between blinded and primed preference test evaluations for each milk type provided relevant, but limited conclusions on the conceptual effect of milk type labeling. However, distinct differences in conceptual patterns were observed for each treatement type when viewed aggregately in the context of stated beliefs/motivations (Figures 1 and 2). For the blinded cohort (Figure 1), differentiation of preference test results and stated motivations behind fluid milk purchase were primarily defined along Factor 1, which accounted for 54.71% of explained variability. In terms of milk type preference, differentiation along Factor 1 indicated clear sensory differences between conventional/locally-farmed milk, which loaded negatively, and organic/pasture-raised milk, which loaded positively. Sensory differences between conventional and pasture-based milk production systems are well-defined in previous literature, with pasture-based systems being defined by higher sweet aromatic flavor, as well as green/grassy and mothball flavors; however, these sensory differences are often unnoticed by consumers (Croissant et al., 2007; Villeneuve et al., 2013). While feed flavors certainly may have played an additive role in milk type differentiation by blind consumers, the conventional/locally-farmed and organic/pasture-raised groupings also differed in thermal processing and packaging materials, which have been cited as significant influences on consumer acceptance (Simon and Hansen, 2001; Lee et al., 2017).

When viewed jointly with self-reported influences/beliefs (CATA), analysis of clustered MCA coordinates from unprimed consumers reinforced these sensory-based groupings and showed that blinded preference patterns in favor of conventional or locally-farmed milk (Cluster 1) were clustered with relatively lower care for price, discounts/sales, brand, product narrative, or indications of farming practices (locally-farmed, family-farmed, validus-certified); however, those who exhibited such patterns were more likely to indicate fat content and package material were important purchase influencers (Figure 1). Alternatively, preference patterns in favor of organic and pasture-raised fluid milks (Cluster 2) were clustered with relatively high care for brand, price, nutritional information, package size, shelf-life, value-added features (rBST-free, DHA-fortified), product narrative, and farming practice attributes (locally-farmed, family-farmed, validus-certified). As expected, blinded preference test evaluations showed inconsistent trends in consumer belief patterns; however, select patterns suggested that subconscious associations between sensory profiles and product attributes may exist. For example, the higher cooked flavor (ultrapasteurized) and similar packaging of organic/pasture-raised milk (cardbord carton) may have proven familiar to organic milk consumers and helped them to align preference with their self-reported motivations, which often include suggestions of ethical consciousness (product narrative, locally-farmed, family-farmed, validus-certified) and better perceived nutrition (Harwood and Drake, 2018; Scozzafava et al., 2020).

Results from primed cohort milk consumers (Figure 2) showed that milk type preference patterns were primarily differentiated along Factor 1, which accounted for 47.62% of explained variability. Differentiation along Factor 1 was defined by separation of locally-farmed/organic (negative loading), pasture-raised (mixed loadings), and conventional (positive loading) milk type groupings, indicating a substantial deviation from the sensory-centric differentiation demonstrated by blind cohort consumers. These results suggest that in the case of fluid milk, attitudinal loyalty generally supercedes sensory preferences, a phenomena noted in other food/beverage studies (Napolitano et al., 2013; Bernard and Liu, 2017). This is further evidenced through alignment of preferences to match self-reported beliefs. Inspection of clustered MCA coordinates revealed that these findings could be defined by three main response clusters.

For primed consumers, preference patterns in favor of locally-farmed/organic (Cluster 1) were associated with relatively higher care for discounts/sales, fat content, nutritional information, value-added features (rBST-free, DHA-fortified), and farming practice attributes (locally-farmed, family-farmed, validus-certified, sustainable practices). These results mirror previous studies that have identified benevolence-driven belief systems and perception of greater product quality as being primary drivers for organic milk purchase (Harwood and Drake, 2018). Furthermore, these results corroborate IAT results of the organic/locally-farmed milk type pairing, indicating that there is indeed overlap in consumer perception regarding locally-farmed and organic milk production systems (Haas et al., 2013). Unlike the first cluster, Cluster 2 was defined by preference patterns against locally-farmed and coincided with higher regard for for organic designation, pasteurization method, and product narrative. As these results directionally trend towards organic milk, but away from locally-farmed, they likely indicate that organic milk consumers uniquely find relative added value in branding/narrative and extended shelf life-features not widely available (extended shelf life) or satisfied (branding tends to be minimalistic for local milk offerings) by locally-farmed products. Previous reasearch on fluid milk consumers has generally concluded that non-conventional fluid milk consumers are more inclined to consider product information and narrative than conventional milk consumers (Getter et al., 2015; Harwood and Drake, 2020). Furthermore, non-conventional consumer subgroups are differentiated by preferences for distinct product attributes (Harwood and Drake, 2020). As such, it is unsurprising that differences exist between organic and locally-farmed milk preference, in terms of underlying belief systems/feature importance. Nevertherless, the relative similarity in consumer perception of organic and locally-farmed, as evidenced in IAT and primed consumer evaluations, indicates that these milk types are largely conceptualized in the same fashion by the majority of consumers.

Finally, Cluster 3 from primed consumers was generally defined by preference patterns in favor of conventional milk and was associated with higher care for price, brand, pasture-raised/grass-fed, package appearance/graphics, package material, and shelf-life. In general, features captured by this cluster closely match sentiments associated with utilitarian milk consumer groups who express a higher purchase likelihood for conventional milk products and high price sensitivity (Rödiger and Hamm., 2015; Harwood and Drake, 2018; Harwood and Drake, 2020). While the third cluster's proclivity towards pasture-raised/grass-fed appears nonsensical, it should be noted that no clear preference pattern was expressed in favor of the pasture-raised milk type designation. Previous studies have shown "grass-fed" to be a universally attractive feature among different milk consumer groups (Harwood and Drake, 2018). Consequently, the present study suggests that the pasture-raised designation for fluid milk may lack consumer-relevant identity relative to other "value-added" milk type designations and may be considered an inherent part of the milk production process, especially when decoupled from branding or package-related information/narrative. This conclusion is further evidenced by IAT exercise findings. Pasture-raised/organic milk type IAT comparisons showed that organic designation, a succinct and certifiable (USDA organic seal) quality indicator for fluid milk, was more often associated with a positive implicit response compared to pasture-raised. On the other hand, local designation, a product feature with inconsistent consumer understanding/beliefs and no formal definition, had no difference in implicit bias when compared to pasture-raised (Hand and Martinez, 2010).

CONCLUSIONS

Implicit association tests from fluid milk consumers revealed implicit biases against conventional fluid milk when compared to organic, locally-farmed, or pasture-raised milk types. Similarly, an implicit bias in favor of organic over pasture-raised milk was reported. Evidence of bias-related influences on preference patterns were noted following primed and unprimed consumer evaluations of fluid milk types. For blinded evaluations, consumers differentiated fluid milk products primarily based upon flavor differences, which can be attributed to dissimilarities in packaging, processing, and feed flavors. Furthermore, there was minimal observed agreement between preference patterns and stated beliefs/purchase influences, indicating fluid milk consumers were largely unable to attribute such flavor distinctions with the associated milk type. On the other hand, preference patterns exhibited in primed evaluations showed product differentiation was primarily based upon agreement with belief systems, indicating that fluid milk consumers are, in general, likely to override sensory preferences to more closely align with stated beliefs. As such, this study serves as evidence that product narrative and consumer perceptions about farming practice are of chief concern for influencing milk type preference, and presumably, purchase. Specifically, findings from this study may be valuable to milk producers of conventional systems. Implicit association testing of fluid milk consumers clearly proves the existence of strong anti-conventional biases. However, specific shortfalls in consumer perception of conventional milk such as animal welfare, sustainability, and lack of product narrative may be improved through alignment with locally-farmed milk marketing/narrative, education initiatives, or general enhancement of product narrative through labeling. Furthermore, the present study may be a valuable resource for milk producers of all types (including those who employ hybrid marketing approaches such as Organic + Pasture-Raised), to review/refine their marketing approach and identify product qualities that may be attractive or alienating to their target consumers.

ACKNOWLEDGEMENTS

Funding was provided in part by The Dairy Alliance (Atlanta, GA) and the National Dairy Council (Rosemont, IL). The use of trade names does not imply endorsement or lack of endorsement of those not mentioned.

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Table 1. Example overview of Implicit Association Test (IAT) structure (Conventional vs. Organic milk example)

Targets	Six (6) photographs of commercial products representing each tested milk type (conventional, organic, locally-farmed, pasture-raised)
Categories	Positive: happy cows, sustainable, healthy, trustworthy, safe, high-quality Negative: unhappy cows, wasteful, unhealthy, misleading, unsafe, low-quality

Block	**No. of Trials**	**Category**	**Left-key Response***	**Right-key Response***
1	20 practice	Target sorting	Conventional Milk Images	Organic Milk Images
2	20 practice	Category sorting	Positive	Negative
3	20 practice	Combined block	Conventional Milk Images + Positive	Organic Milk Images + Negative
4	40 collected	Combined block	Conventional Milk Images + Positive	Organic Milk Images + Negative
5	40 practice**	Category sorting	Organic Milk Images	Conventional Milk Images
6	20 practice	Combined block	Organic Milk Images + Positive	Conventional Milk Images + Negative
7	40 collected	Combined block	Organic Milk Images + Positive	Conventional Milk Images + Negative

*Presentation of targets and categories was balanced for position among participants (Nosek et al., 2005)

**Lengthened Block 5 practice set is intended to clear learned behavior based on positioning of earlier blocks (Nosek et al., 2005)

Table 2. Implicit Association Test (IAT) scores for fluid milk type pairings

	Comparison	Conventional (+) vs. Organic (-)	Conventional (+) vs. Locally-Farmed (-)	Conventional (+) vs. Pasture-Raised (-)	Organic (+) vs. Locally-Farmed (-)	Organic (+) vs. Pasture-Raised (-)	Pasture-Raised (+) vs. Locally-Farmed (-)
Overall Test Statistics	Total N	253	249	254	239	249	237
	Valid N*	232	226	237	216	229	216
	Mean D-score**	-0.26	-0.28	-0.23	-0.02	0.10	-0.03
	D-score SD	0.55	0.58	0.59	0.52	0.44	0.53
	*P****	**< 0.001**	**< 0.001**	**< 0.001**	0.514	**0.001**	0.409
	Effect Size (*d*)	-0.46	-0.48	-0.38	-0.04	0.22	-0.06
Implicit Association Strength Classifications**	Strong (-)	25.43%	26.55%	32.07%	12.04%	6.11%	11.11%
	Moderate (-)	21.12%	20.35%	11.39%	18.06%	7.86%	15.74%
	Slight (-)	11.21%	12.39%	12.24%	12.04%	13.10%	16.67%
	Neutral	16.38%	17.70%	16.88%	21.76%	22.71%	18.98%
	Slight (+)	10.34%	6.19%	9.28%	11.57%	22.71%	16.67%
	Moderate (+)	11.21%	10.18%	10.97%	15.28%	19.65%	9.26%
	Strong (+)	4.31%	6.64%	7.17%	9.26%	7.86%	11.57%

*Valid N refers to number of responses used to calculate test statistics after dropping incomplete responses and responses with excessive completion speed

**Implicit association strength classifications follow conventional guidelines for calculating and classifying individual D-scores- a standardized measure of implicit bias (Greenwald et al., 2003; Rooth, 2010). A negative D-score indicates a bias directionally in favor of the negative (-) designated milk type, and a positive D-score indicates a bias directionally in favor of the positive (+) designated milk type. These positive/negative designations can be noted in the first row of the above table.

***Bolded p-values indicate significance

Table 3. Summary of milk type preference test results from fluid milk consumers (n=174 total)

Comparison	**Treatment**	**Panelists**	**2-AFC Results**	***p(Binomial)***	***p* (χ,^{2})**
Conventional vs. Organic	Blind	87	Conventional- 31, Organic- 56	**0.010**	0.436
	Primed	87	Conventional- 36, Organic- 51	0.133	
Conventional vs. Locally-Farmed	Blind	87	Conventional- 45, Locally-Famed- 42	0.830	0.094
	Primed	87	Conventional- 34, Locally-Farmed- 53	0.053	
Conventional vs. Pasture-Raised	Blind	87	Conventional- 51, Pasture-Raised- 36	0.133	0.171
	Primed	87	Conventional- 42, Pasture-Raised-45	0.830	
Organic vs. Locally-Farmed	Blind	87	Organic- 49, Locally-Farmed- 38	0.284	0.129
	Primed	87	Organic- 39, Locally-Farmed- 48	0.391	
Organic vs. Pasture-Raised	Blind	87	Organic- 50, Pasture-Raised- 37	0.198	0.878
	Primed	87	Organic- 51, Pasture-Raised- 36	0.133	
Pasture-Raised vs. Locally-Farmed	Blind	87	Pasture-Raised- 26, Locally-Farmed- 61	**<0.001**	0.113
	Primed	87	Pasture-Raised-36, Locally-Farmed- 51	0.133	

*Bolded p-values indicate significance (α=0.05)

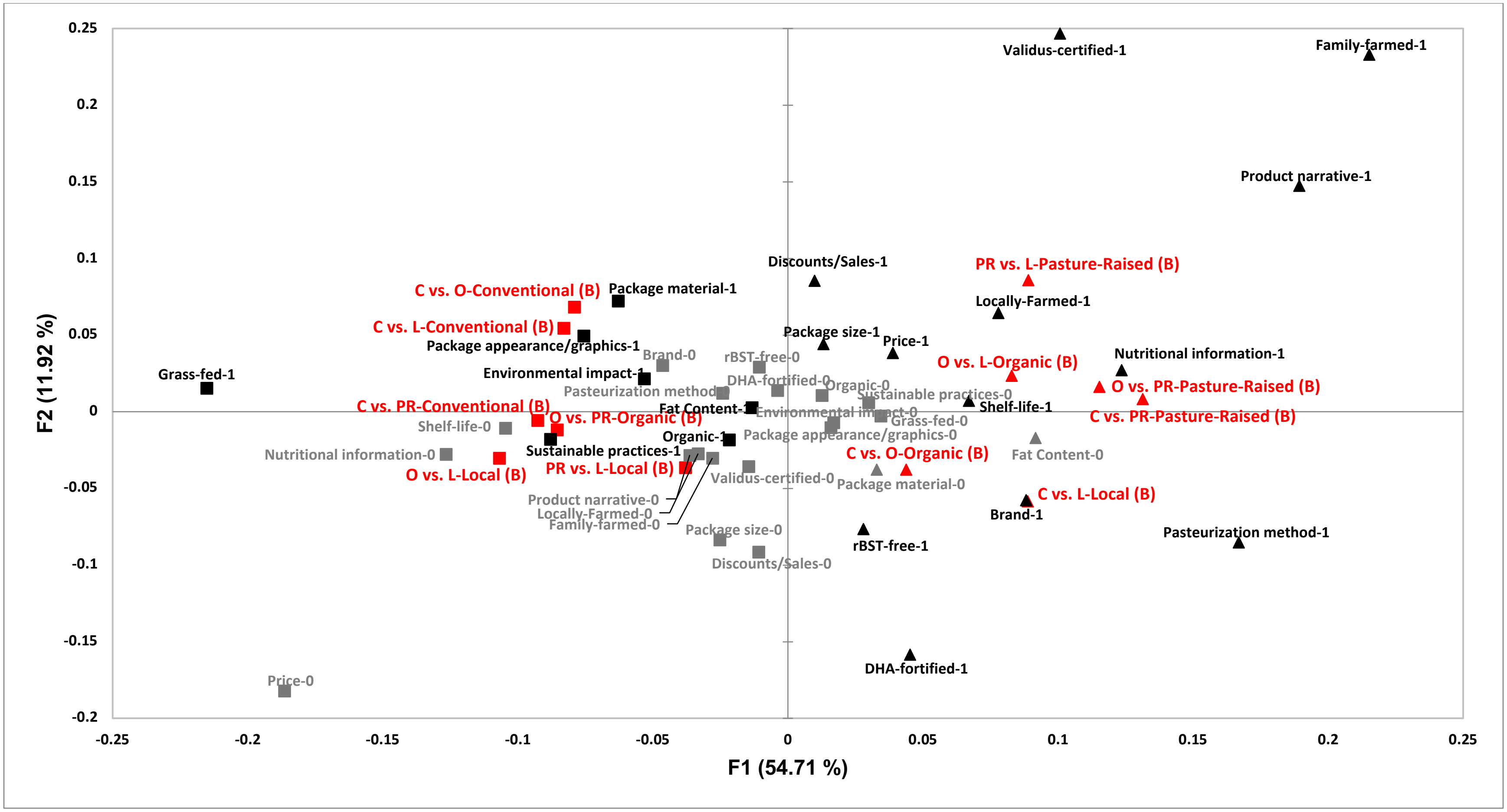

Figure 1. Multiple correspondence analysis of blinded cohort (n=87) preference test results and self-reported influences on fluid milk purchase

*C=conventional, O=organic, L=locally-farmed, PR=pasture-raised; B=blinded; preference test results are listed in the following order: pairing type, preference choice, priming treatment

**1=attribute was chosen in check-all-that-apply (CATA) exercise, 0=attribute was not chosen in CATA exercise

***■=Cluster 1 preferences/purchase influences, ▲=Cluster 2 preferences/purchase influences

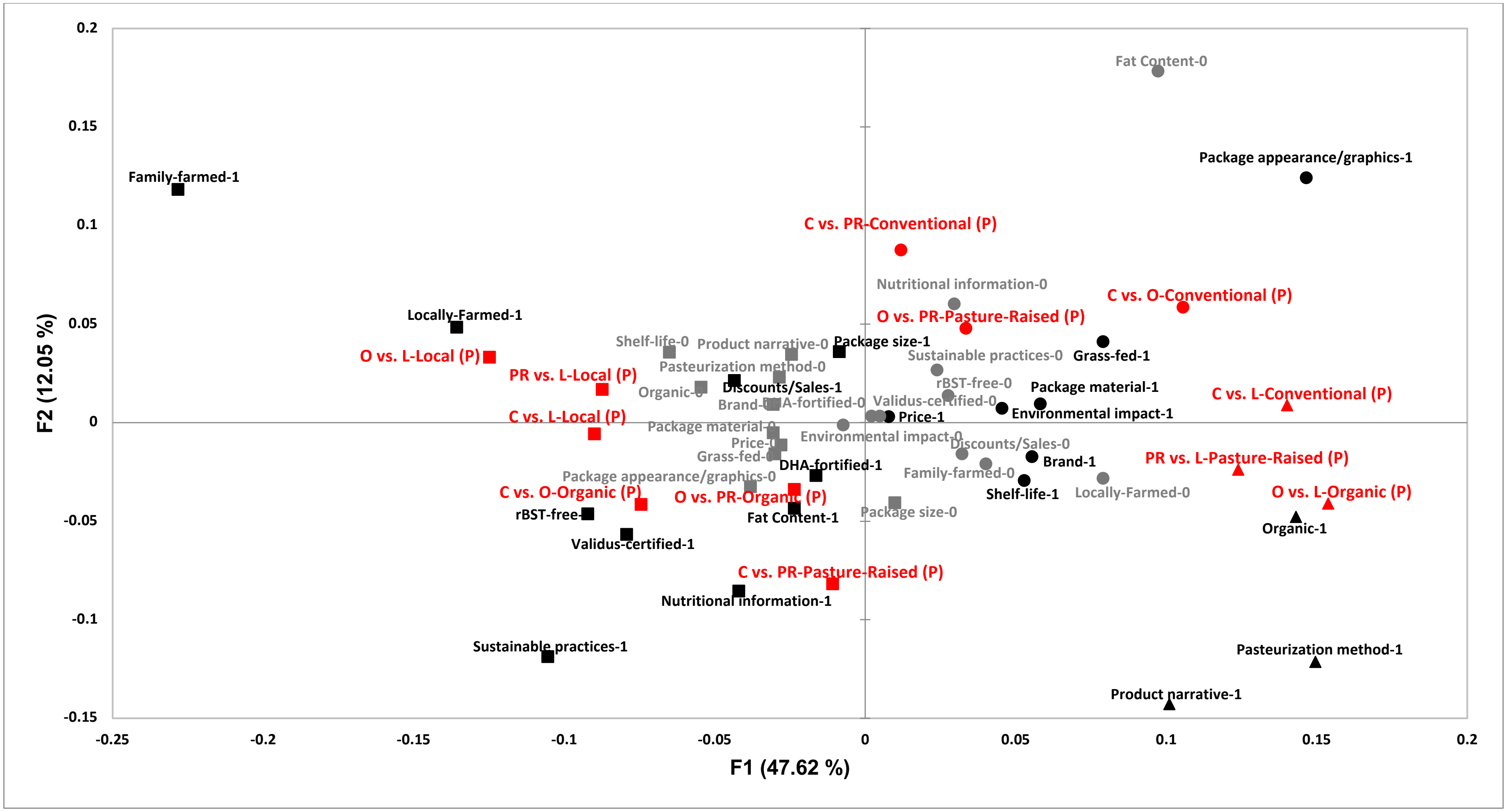

Figure 2. Multiple correspondence analysis of primed cohort (n=87) preference test results and self-reported influences on fluid milk purchase

*C=conventional, O=organic, L=locally-farmed, PR=pasture-raised; P=primed; preference test results are listed in the following order: pairing type, preference choice, priming treatment

**1=attribute was chosen in check-all-that-apply (CATA) exercise, 0=attribute was not chosen in CATA exercise

***■=Cluster 1 preferences/purchase influences, ▲=Cluster 2 preferences/purchase influences, ●= Cluster 3 preferences/purchase influences

CHAPTER 5: Application of Temporal Penalty Analysis for the Optimization of Sugar Reduction in Protein Beverages

Application of Temporal Penalty Analysis for the Optimization of Sugar Reduction in Protein Beverages

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The content of this chapter was submitted for publication in:

Journal of Sensory Studies (in review)

ABSTRACT

Penalty analysis is a well-accepted and integral part of new product development and product optimization. This study evaluated the application of penalty analysis to temporal evaluation of vanilla-flavored protein beverages sweetened with different sweeteners to elicit temporal drivers of liking and disliking. Iso-sweet concentrations of sucrose, sucralose, stevia, and monk fruit, as well as natural sweetener blends, were assessed in the study. Ready-to-mix (RTM) protein beverages were formulated with flavoring, sweetener, and whey protein isolate (25 g protein per 360 mL serving). Untrained consumers ($n=143$ total) were recruited to participate in both temporal liking and temporal check-all-that-apply (TCATA) exercises. Attributes evaluated during TCATA were cardboard and vanilla flavors, sweet taste, bitter taste, metallic, and astringency. Consumer results from temporal liking were matched at each second with results from their TCATA profiling. Significant penalties to liking associated with the presence or absence of attributes were subsequently identified. The sucrose formulation received no penalties and had the most consistent temporal liking of all formulations. Monk fruit beverages were penalized ($p<0.05$) for metallic sensation over the course of aftertaste evaluation, and stevia beverages were penalized for bitter and metallic tastes, astringency, and lingering bitterness. Natural sweetener blends proved effective at mitigation of these unpleasant temporal sensory qualities. Understanding the temporal aspects of protein beverages related to the different sweetener types may provide guidance for formulating reduced-sugar products that maintain an acceptable consumer temporal experience.

PRACTICAL APPLICATION

There is ample consumer demand for high-protein products without added sugar. However, nonnutritive sweetener systems are often less palatable than sucrose-sweetened products, presenting a barrier to more widespread consumption. This study utilized a penalty analysis approach from a temporal perspective to identify formulations that are viable alternatives to nutritive sweetener formulations. Findings from this study may be of specific interest to manufacturers of high-protein products. Additionally, the approaches used within this study may hold practical value for food manufacturers as a whole, as they outline a novel consumer-centric approach for refining product formulations.

Key Words: Temporal Check-All-That-Apply, TCATA, Penalty Analysis

INTRODUCTION

Increased protein consumption has been a powerful trend among consumers in both developed and developing countries consumers over the last few decades (Henchion, Hayes, Mullen, Fenelon, & Tiwari, 2017). With increased demand for high-protein products, whey protein powders and beverages present options for consumers that are convenient, nutritious, and available in a wide variety of flavors (Rittmani, 2006).While ready-to-mix protein powders and ready-to-drink protein beverages have traditionally been viewed as products specific to sport nutrition and recovery, this increased interest has expanded to various other consumer groups. Aside from workout recovery, high-protein products are consumed for several purposes, including meal replacement, nutritional supplementation, and maintenance of satiety (Harwood & Drake, 2018). Along with increased protein consumption, concerns related to sugar reduction and the use of natural ingredients are often mentioned as unifying features of health-consciousness. Previous research has demonstrated the importance of naturally sweetened protein beverages with low carbohydrate contents to consumers (Oltman, Lopetcharat, Bastian, & Drake, 2015). While conceptually appealing, natural non-nutritive sweeteners are often associated with decreased consumer acceptance due to the presence of off-flavors and differences in temporality compared to sucrose (Li, Lopetcharat, & Drake, 2015; Tan, Wee, Tomic, & Forde, 2019). Previous studies have suggested that these sensory deficiencies may be addressed through the formulation of blends or by partial (in place of complete) sugar-reduction approaches (Parker, Lopetcharat, & Drake, 2018). However, the consumer-relevant impact of such approaches to assuage temporality concerns has not been adequately addressed, necessitating further evaluation of natural non-nutritive sweeteners and natural sweetener blends in common commercial applications (such as protein beverages).

Temporal (time-dependent) sensory evaluation techniques are often employed to track the dynamic experiences inherent to food and beverage consumption. Early temporal studies were generally oriented at the evolution of basic taste intensity over time, a method known as time-intensity (TI) (Cliff & Heymann, 1993). Experiments by Taylor and Pangborn (1990) posited that TI methodologies could similarly be employed to track hedonic measures of food and beverage consumption. As technological advances have facilitated the ease of use for temporal collection and subsequent analyses of temporal data, several advantages have been identified. For example, findings from Thomas et al. (2015) have reinforced the value of temporal liking (TL), as it produced more discriminating results than traditional static measurements of liking with a 9-pt hedonic scale.

Among the more recently developed temporal sensory techniques, temporal check-all-that-apply (TCATA) has established itself as a useful procedure for profiling complex food and beverage products (Ares et al., 2015; Castua, Antúnez, Giménez, & Ares, 2016). As the name suggests, TCATA is a temporal extension of static check-all-that-apply (CATA) questions, wherein a panelist may select multiple choice options at once to respond to a question. Within a TCATA project, panelists similarly identify the presence of all present attributes in a product, but do so over time by selecting and deselecting predetermined attribute options over the course of evaluation (Alcaire et al., 2017). In turn, the evolution of sensory perception for a product may be mapped based on aggregated results, reported as the proportion of the population who selected a given product attribute at a given time. Compared to other common multivariate temporal analyses, such as temporal dominance of sensations (TDS), TCATA has been shown to provide better discriminatory power (Berget, Castura, Ares, Næs, & Varela, 2020). TCATA evaluations have been performed on multiple product categories, with both trained and untrained assessors (Ares et al., 2016; Jaeger et al., 2017; Harwood, Parker, & Drake, 2020). Additionally, TCATA has proven effective in the temporal profiling of various sweeteners when evaluated on their own (Reyes, Casutra, & Hayes, 2017; Tan, Wee, Tomic, & Forde, 2019), or in beverage applications (Ares et al., 2015; Parker, Lopetcharat, & Drake, 2018). In terms of exercise execution, there is a general agreement that studies with trained assessors unsurprisingly provide better sample discrimination than untrained consumers; however, these effects may be mitigated for untrained consumers through the employment of manageable attribute list sizes and familiarization with the TCATA task (Jaeger et al., 2017; Jaeger et al., 2018; Rizo, Vidák, Fiszman, & Tarrega, 2020).

In static sensory evaluations, the combination of liking scores and consumer-derived sensory assessment of select product attributes is often used to conduct penalty (mean drop) analyses. Penalty analysis is a product optimization technique that relates acceptance and attribute ratings on an individual level to directionally guide product reformulation (Lawless & Heymann, 2010). Traditionally, penalty analysis is conducted using just-about-right (JAR) scores to identify attribute-specific deviations from sensory ideals (Lawless & Heymann, 2010); however, the application of penalty analysis to CATA data has also proven useful for identifying drivers of liking and opportunities for reformulation for food/beverage products (Ares, Dauber, Fernández, Giménez, & Varela, 2014). Findings by Ares et al. (2017) showed that JAR and CATA-based penalty analyses resulted in largely similar findings, although JAR scaling identified more significant penalties than CATA overall.

In alignment with the aim of penalty analysis to join consumer acceptance and descriptive attribute information, combinatory temporal techniques have been proposed in recent years for the purposes of defining the consumer temporal experience and identifying drivers of liking.

Thomas, Visalli, Cordelle, & Schlich (2015) were among the first to investigate temporal drivers of liking through the combination of TDS and TL evaluations (applied in separate sessions) on French cheeses. Findings from this study served as definitive proof that product acceptance may be influenced by specific temporal sensory attributes. Subsequent studies by Thomas et al. (2017) further proposed that TDS and TL could be administered simultaneously on a single interface to identify temporal drivers of liking, although the use of such methods with untrained consumers prompted concerns related to ease-of-use. As it applies to TCATA data, investigation of the relationship between temporal profiles and liking has generally been approached using static measurements of overall liking, or a "before and after" liking approach (Ares et al., 2017; Palgarini et al., 2020). Additionally, Ramsey et al. (2020) have recently suggested using a combined TL/TCATA approach to identify specific time segments that dictate overall liking in beers. However, no studies, to our knowledge have combined TCATA and TL data streams second-for-second to understand potential temporal drivers of liking at specific times.

The application of natural sweetener systems in beverages has previously been studied in a manner that addressed consumer acceptance and temporal profiles separately. However, meaningful insights associated with product formulation could be gained from the combination of these data streams. The present study proposes that the joint application of TCATA and TL may be analyzed with traditional penalty analysis techniques to provide consumer-relevant insights into the temporal effects of protein beverage formulation and selection of sweetener system. Generation of such data may assist product development initiatives by helping to identify specific areas where products or ingredient performance can be improved from a temporal perspective.

MATERIALS AND METHODS

Sample Formulation and Preparation

Protein beverages (25 g protein/360 mL water) were formulated using whey protein isolate (Provon 192; Glanbia Nutritionals, Twin Falls, ID), deionized water (DIW), and vanilla flavorings (natural vanilla 1032, natural cream 0151, natural French vanilla 1068; Flavor Artistry, Corona, CA) (Table 1). Additionally, protein beverages were either individually sweetened with iso-sweet concentrations of crystalline fructose (Krystar 300; Tate & Lyle, Decatur, IL), sucrose (Imperial Sugar Co., Sugar Land, TX), sucralose (Hard Eight Nutrition, Henderson, NV), monk fruit extract (Purefruit Select monk fruit extract; Tate & Lyle), stevia leaf extract (Tasteva stevia sweetener; Tate & Lyle), or natural sweetener blends (fructose, stevia, monk fruit), as described by Parker, Lopetcharat, & Drake (2018) (Table 1). Pre-weighed dry mixes of these ingredients were mixed in batches with 500 mL of DIW 15 minutes prior to serving using shaker bottles with metal blender balls (BlenderBottle, Lehi, UT).

Temporal Penalty Analysis of Protein Beverages

Temporal penalty analysis (TPA) is a joint analysis of data from both temporal liking (TL) and temporal check-all-that-apply (TCATA) methodologies. Data collection for TPA was conducted over two 2-day sessions (4 days total). Protein beverages (Table 1) with individual sweeteners (fructose training sample, sucrose, sucralose, stevia, monk fruit) were evaluated in the first session, and protein beverages with reduced-sugar sweetener blends (fructose training sample, 50% fructose/50% stevia, 50% fructose/50% monk fruit, 50% fructose/25% stevia/25% monk fruit, 25% stevia/75% monk fruit) were evaluated in the second session. Within each 2-day session, day 1 consisted of TL profiling and day 2 consisted of TCATA profiling of each beverage. The decision to keep TL fixed on day 1 of testing for both sessions was to deter influence on acceptance that may be gained from increased knowledge of product sensory features during TCATA evaluation (Popper, Rosenstock, Schraidt, & Kroll, 2004). Untrained panelists were recruited for each session (n=71 session 1, n=72 session 2) from the North Carolina State University Sensory Service Center database (Raleigh, NC). In order to qualify for participation, panelists were required to self-report frequent (once a week or more often) consumption of protein beverages, frequent (once a week or more often) exercise habits, purchase of ready-to-mix protein powder within the last month, and acceptance for vanilla-flavored protein beverages. Panelists were compensated for full participation in the study with a $10 gift card to a local store.

Upon arrival for testing, panelists were given a brief instruction on how to evaluate the products using TL or TCATA methodologies. Test instruction included a brief demonstration by the lead researcher, on how to use the software on an iPad interface and included a demonstration of notable time points (expectoration, evaluation duration) to be aware of. Following instruction, a single practice sample (fructose-sweetened beverage) was presented to the panelists so they could further familiarize themselves with the iPad interface/software (Compusense Cloud, Guelph, ON) and, in the case of TCATA evaluation, the pertinent sensory attributes being tested. Continuous measurement of temporal liking was completed using a 9-pt scale, labeled with the following anchors: 1=Dislike Extremely, 5=Neither Like nor Dislike, and 9=Like Extremely. At the beginning of evaluation, liking scores were fixed at a score of 5, but were able to be changed immediately after beginning the evaluation. Attributes evaluated using TCATA included cardboard and sweet aromatic/vanilla flavors, sweet and bitter tastes, astringent and metallic (Parker et al., 2018) and no limits were placed on concurrent selections, or duration of selection (no fading; Ares et al., 2016). A brief explanation of each sensory attribute was also provided prior to evaluation of the practice sample (Table 2).

Following test instruction and completion of the practice beverage, panelists were presented with 4 samples of vanilla-flavored protein beverages, one at a time, in a fully randomized design, counterbalanced for position effects (William's design). Batches (500 mL) of each sample were prepared using chilled (4°C) deionized water approximately 15 minutes prior to serving, as previously described, and were then stored at refrigerated temperature (4°C) until serving. Protein beverage samples were given to panelists in 20 ml aliquots in lidded 60 mL soufflé cups labeled with 3-digit blinding codes. Sample evaluation consisted of in-mouth and aftertaste stages, with a total evaluation time of 195 s. Once presented with a sample, panelists were instructed remove the lid before proceeding to take the entire sample (20 mL) in their mouth and start the temporal evaluation. Panelists temporally profiled (TL or TCATA) the product in-mouth for 15 s, at which point they were prompted to expectorate the sample by an on-screen cue. Following expectoration, panelists continued to profile the aftertaste profile of the sample in the same manner for 180 s until an elapsed time of 195 s was completed. Following sample evaluation, there was a 4-min enforced rest period before the next sample was served, during which time panelists were encouraged to cleanse their palates using available unsalted crackers and deionized water. This process was repeated by each panelist for 4 sample evaluations per day, on both days of testing (TL and TCATA). All aforementioned serving, consumer evaluation, and rest steps were consistent across both singular sweetener (session 1) and blend (session 2) sessions.

Statistical Analysis

Following data collection (Compusense Cloud, Guelph, ON), smoothing of TL and TCATA curves was conducted in "tempR" package in R version 3.3.1 (Castura, Antúnez, Giménez, & Ares, 2016). Smoothed TL and TCATA profile data was used to identify relevant temporal metrics, such as area under the curve (AUC). Additionally, peak citation proportion and time to peak citation proportion (T_{max}) were identified for each TCATA attribute. AUC, peak citation proportion, and T_{max} values were subsequently subjected to agglomerative hierarchical clustering (AHC) using a dissimilarity matrix with Euclidean distance proximity type, Ward's agglomeration method, and entropy-based truncation in order to understand overarching formulation similarities/differences. In order to identify temporal penalties, smoothed temporal data was exported and arranged so as to match up TL and TCATA data second-for-second for each individual consumer. Analysis of significant ($p<0.05$) penalties was then conducted for each sample using the penalty analysis module in XLSTAT version 2019.3.2.61685 (Addinsoft, New York, NY). Within the penalty analysis routine, mean liking scores were compared between assessors who indicated presence and assessors who indicated absence of a given attribute at each second of evaluation. Penalties were identified if the proportion of assessors who indicated the presence of an attribute exceeded 20%, and if the mean liking score of that group differed significantly (Fisher's LSD, $\alpha=0.05$) from that of assessors who did not report the attribute's presence. Following penalty analysis, smoothed TL/TCATA data was graphed on a 2-axis scatter plot, with the left axis representing citation proportions from the TCATA exercise, and the right axis representing overall liking score (TL) during consumption. Red highlights were superimposed on TCATA curve regions where penalties ($p<0.05$) were assessed for the presence of a given attribute, and green highlights were superimposed on curve regions where significant ($p<0.05$) lifts in liking were noted for the presence of a given attribute (Figures 1 and 2).

RESULTS AND DISCUSSION

Session 1: Individual Sweetener Formulations

Evaluation of TCATA profiles for vanilla protein beverages with individual sweeteners were largely in agreement with findings from previous studies (Parker, Lopetcharat, & Drake, 2018; Tan, Wee, Tomic, & Forde, 2019) (Table 3, Figure 1). Analysis of area under the curve (AUC) for various attributes in the TCATA exercise showed that perception was largely characterized by sweet taste and sweet aromatic/vanilla flavor, which were readily apparent in-mouth and extended through the aftertaste for each formulation. However, formulations were primarily differentiated by bitter taste, cardboard flavor, and metallic flavor perception, as evidenced by their relatively higher standardized ranges of AUC values (Range/Max). Additionally, astringency, cardboard flavor, metallic, and sweet taste were the most variable in terms of time to peak citation proportion, reinforcing that different sweeteners imparted varied temporal evolution characteristics in addition to varied attribute intensities.

The sucrose-sweetened formulation was characterized by lower AUC and peak citation rate, and earlier onset for astringency and sweet aromatic/vanilla, compared to other individual sweetener formulations. Furthermore, the sucrose formulation exhibited the lowest peak citation rate, and fastest curve decay for sweet taste among session 1 formulations (Figure 1), in agreement with TCATA curves reported by Reyes, Castura, and Hayes (2017). Interestingly, the sucrose formulation was profiled as having both bitter taste and metallic flavor, though trained descriptive panels have routinely reported sucrose sweetened protein beverages to be devoid of these sensory attributes (Parker, Lopetcharat, & Drake, 2018). Citation of bitter taste and metallic flavor were similarly documented in TCATA evaluations of sucrose by Tan, Wee, Tomic, and Forde (2019), suggesting that citation of these features can be attributed to user error or mischaracterization by a subset of consumers. Overall, sucralose performed more similarly to sucrose than stevia or monk fruit, as evidenced by cluster analysis (Figure 3). Stevia and monk fruit, on the other hand, were defined by relatively higher AUC values for bitter taste, metallic flavor, and astringency and were grouped as dissimilar to sucrose and sucralose sweetened beverages.

The evolution of liking over the course of product evaluation was also assessed for each individual sweetener system. Overall, the sucrose-sweetened protein beverage exhibited an increasing liking trend during in-mouth evaluation and received characteristically consistent liking ratings throughout aftertaste evaluation. Similarly, the TL curve of the sucralose-sweetened formulation was characterized by relatively uniform liking scores throughout evaluation, although liking magnitude was slightly lower throughout evaluation compared to sucrose. Conversely, monk fruit and stevia-sweetened formulations were characterized by decreased liking trends in-mouth, followed by recovery post-expectoration.

While viewing TL and TCATA data streams separately provides useful insights on a comparative basis, the application of TPA to temporal data streams successfully identified consumer-relevant shortcomings in product performance related to different sweeteners that were not readily apparent (Figure 1). As previously mentioned, the TL curve for the sucrose-sweetened formulation was characterized by relatively higher and consistent liking scores throughout evaluation. Analysis of the sucrose formulation temporal profile with the application of TPA confirmed that this profile was not only relatively more liked, but that the temporal profile of the sucrose-sweetened vanilla protein beverage formulation was devoid of penalties over the course of consumption. A brief lift ($p<0.05$) in liking associated with sweet taste perception from 39s to 42s was additionally noted. Similarly, the sucralose-sweetened formulation received no significant penalties during evaluation, but did have a lift ($p<0.05$) attributed to sweet aromatic/vanilla flavor in-mouth from 7s to 12s. Temporal sensory and hedonic similarities between sucrose and sucralose are well-cited (Zorn, Alcaire, Vidal, Giménez, & Ares, 2014; Rocha & Bolini, 2015), corroborating the validity of TPA findings. However, negative conceptual perception of sucralose as an artificial sweetener necessitates exploration of natural sweeteners that can perform akin to sucrose in beverages (Parker, Lopetcharat, & Drake, 2018).

Natural nonnutritive stevia and monk fruit formulations exhibited temporal penalties ($p<0.05$) during both in-mouth and aftertaste evaluation, coinciding with their aforementioned periods of decreased liking and recovery. The stevia-sweetened formulation received significant penalties ($p<0.05$) for metallic and bitter tastes in-mouth which extended into early aftertaste. Additionally, this formulation received a prolonged on-and-off penalty ($p<0.05$) associated with astringent mouthfeel from 25s to 118s. Interestingly, the citation proportion for astringency in the stevia formulation did not significantly differ from other formulations (results not shown). This result suggests consumer misunderstanding related to attribute identification/selection and may indicate consumer confusion of astringency with bitter taste or off-flavor, as reported in previous studies (Lee & Lawless, 1991). While consumer misidentification is a concern that should be considered and mitigated when possible, the power of TPA to identify lapses in consumer acceptance despite decreased attribute understanding should be noted as a distinct advantage of this analysis approach. The monk fruit beverage was, overall, less penalized than the stevia formulation, but received penalties in the aftertaste associated with metallic (22s to

34s, 77s to 107s) and sweet aromatic flavor (38s to 41s, 46s to 51s). Evaluation of temporal profiles and penalties indicates that stevia and monk fruit are indeed characterized by the presence of off-flavors, and that the presence of these flavors negatively impacts consumer acceptance. Furthermore, the evolution of dominant flavors in protein solutions sweetened with stevia and monk fruit have been reported to be relatively similar (Wagoner, McCain, Foegeding, & Drake, 2018). However, the present study noted that subtle differences in temporal presence (and, presumably, intensity) of certain attributes may result in different penalty profiles. In particular, stevia was characterized by comparatively earlier penalties, while monk fruit was defined by penalized attribute presence later in the aftertaste. These findings are consistent with time intensity results reported by Parker, Lopetcharat, and Drake (2018), who showed stevia was defined by an earlier, steeper, and more intense bitter taste curve than monk fruit. The presence of distinct sensory penalties with different nonnutritive sweeteners presents the possibility that sweeteners with different temporal profiles may be combined to avoid harsh off-flavor perception and the corresponding decrease in consumer acceptance.

Session 2: Natural Sweetener Blend Formulations

As suggested by findings from Parker, Lopetcharat, and Drake (2018), amelioration of product performance concerns and achievement of sugar reduction for naturally sweetened protein beverages may be realized through the application of sweetener blends. The application of TPA showed that sugar-reduced sweetener blend formulations, such as 50% fructose/50% stevia and 50% fructose/50% monk fruit received fewer penalties than their nonnutritive counterparts (100% stevia or 100% monk fruit). The 50% fructose/50% stevia formulation still received brief penalties related to bitter taste (24-35s), metallic flavor (56-68s), and astringency (~102-126s) in the aftertaste, but penalties related to off-flavors in-mouth were largely avoided.

The 50% fructose/50% monk fruit formulation was similarly characterized by only brief penalties in the early aftertaste related to astringency (29-36s), metallic flavor (31-36s) and bitter taste (26-27s, 37-38s). In both formulations, using a sugar-reduced approach rather than a 100% natural nonnutritive sweetener system resulted in improved product performance and avoided sustained attenuations in consumer acceptance. Overall, partial replacement of nonnutritive sweeteners with fructose resulted in temporal profiles that were clustered as relatively more similar to sucrose or sucralose, than to 100% stevia or monk fruit (Figure 3). Furthermore, the avoidance of prolonged decreases in temporal liking suggests that consumer thresholds for off-flavor intensity were not met as readily in sugar-reduced protein beverage formulations.

For formulations that included a blend of both stevia and monk fruit, TPA revealed a synergistic effect from complementary temporal profiles. Within the 50% fructose/25% stevia/25% monk fruit formulation, marked reductions in penalties were noted compared to other sugar-reduced formulations- an indication of overall product performance and consumer acceptance improvement that agrees with findings from Parker, Lopetcharat, and Drake (2018). Similar synergistic effects were also noted by Hanger, Lotz, and Lepeniotis (1996), who reported that select blends of high intensity sweeteners more closely matched the flavor profile of sucrose than any singular sweetener using trained sensory profiling techniques. The complimentary effects of monk fruit and stevia were also demonstrated for the nonnutritive 25% stevia/75% monk fruit blend formulation. Compared to 100% stevia or 100% monk fruit formulations, the 25% stevia/75% monk fruit blend exhibited only brief penalized periods for cardboard (50s to 52s) and lingering sweet taste (142s to 192s). No in-mouth penalties due to bitter taste or metallic flavor were found. Furthermore, liking was generally consistent throughout evaluation, and was free from the reduction concurrent with in-mouth evaluation of other natural nonnutritive formulations.

Temporal Penalty Analysis Advantages

Overall, the practical value of temporal penalty analysis as a means for identifying lapses in product performance and addressing reformulation initiatives was apparent in the present study. In addition to garnering reformulation insights, the employment of TPA in the present study highlights several features that make it a useful sensory analysis tool. In particular, TPA is characteristically consumer-centric in its combinatorial use of TL and TCATA methods. Collection of temporal sensory profiles with similar methodologies such as TDS is, by comparison, less conducive to success with untrained consumers given the ambiguity of "dominance" (Varela et al., 2017) and higher tedium (Ares et al., 2015). As previously mentioned, TL and TCATA also present advantages in discriminatory ability compared to alternative methods, both static (Thomas et al., 2015) and temporal (Berget, Castura, Ares, Næs, & Varela, 2020).

From an analysis perspective, the application of penalty analysis to temporal data streams is similarly expedient. Existing analysis methods for TCATA data generally include visual investigation of curve morphology, key metrics (T_{max}, AUC, etc.), and generation of difference curves for sample comparison (Castura, Antúnez, Giménez, & Ares, 2016; Tan, Wee, Tomic, & Forde, 2019). While attention to these findings are important to understanding temporal product experiences, the practical relevance to product performance is often dependent on comparison to a "gold standard". In the present study, the sucrose formulation would likely be considered the standard for performance, and success would be measured by similarity to the sucrose formulation temporal profile. However, in the scope of new product development and product optimization campaigns, an appropriate standard may not exist. Because TPA presents temporal sensory profiles in the context of consumer acceptance the necessity of comparison-based methods is removed. Furthermore, the resulting TPA curves are precise (to the second), are able to stand alone, and insights can be readily communicated using penalty-analysis terminology that is widely understood in the sensory and product development fields (Rothman, 2007). Future studies should be conducted to understand the performance of TPA as applied to various food/beverage applications, consumption styles (swallowing time, multi-sip, mastication) (Galmarini, Loiseau, Visalli, and Schlich, 2016; Jourdren et al., 2016), and test parameters (attribute list length, simultaneous data stream capture) (Jaeger et al., 2018; Thomas et al., 2017), as these concepts were not adequately explored in the present work. However, the results of this study suggest merit in the approach and results that may be obtained through temporal penalty analysis and warrant its consideration for future temporal studies.

CONCLUSIONS

This study proposed the application of temporal penalty analysis as a means for identifying sensory shortcomings in vanilla protein beverages from a temporal perspective and directing reformulation opportunities. TPA showed clearly that consumer acceptance of 100% stevia and 100% monk fruit formulations suffered from lingering bitter and metallic off-flavors which were apparent to consumers in-mouth and extended into aftertaste perception. Furthermore, it was readily apparent that 100% sucrose and 100% sucralose formulations did not suffer the same temporal penalties to acceptance, as these flavors were not cited, or were not cited at critical levels. As sugar-reduced, sugar-free, and natural sweetener systems are conceptually attractive and conducive to a healthful lifestyle, natural sugar-reduced and natural nonnutritive sweetener blends were also assessed. Sugar-reduced blends showed comparative advantages to 100% natural nonnutritive (stevia, monk fruit) sweetener systems, as evidenced by the amelioration of temporal penalties. Specifically, the 50% fructose/25% stevia/25% monk fruit formulation showed almost full clearing of temporal penalties, suggesting multiple sweeteners blends may provide complementary temporal effects. Similarly, beverages with a blend of 25% stevia/75% monk fruit showed marked improvement in consumer-relevant product performance compared to beverages with singular natural nonnutritive sweeteners (100% stevia or 100% monk fruit). The blending of the two sweeteners at these ratios avoided bitter/metallic penalties and the associated decrease in consumer acceptance following in-mouth evaluation, although penalties related to lingering metallic flavor remained a concern.

Aside from the practical findings related to protein beverage formulations and the application of different sweetener systems, temporal penalty analysis was an effective addition to existing temporal analysis methods. One of the primary advantages of TPA is the utilization of consumer-centric TL and TCATA exercises, which are easily understood and facilitate effective product discrimination with untrained assessors. Furthermore, the results from TPA are comprehensible, communicable, precise (to the second) and stand alone without the requirement of between-product comparisons. Additional studies should be conducted to confirm the widespread application of TPA for foods and beverages of various complexities. TPA should be considered as an effective new tool in the ever-growing library of temporal sensory methodologies.

ACKNOWLEDGEMENTS

Funding provided in part by the National Dairy Council (Rosemont, IL). The use of tradenames does not imply endorsement nor lack of endorsement by those not mentioned.

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Table 1. Formulations for sweetened vanilla ready-to-mix whey protein beverages at a concentration of 25g protein per 12 oz (360 mL) deionized water

	Sample	**Fructose (g/L)**	**Sucrose (g/L)**	**Sucralose (mg/L)**	**Monk fruit (mg/L)**	**Stevia (mg/L)**	**Vanilla Flavor (g)**	**Cream Flavor (g)**	**French Vanilla Flavor (g)**	**Protein (g)**	**Water (mL)**
Individual Sweeteners (Session 1)	Fructose (Control)	31.80					0.83	0.63	0.21	38.92	500
	MF				321.32		0.83	0.63	0.21	38.92	500
	Stevia					393.26	0.83	0.63	0.21	38.92	500
	Sucralose			65.36			0.83	0.63	0.21	38.92	500
	Sucrose		35.00				0.83	0.63	0.21	38.92	500
Sweetener Blends (Session 2)	FRU50/ STV50	13.82				139.49	0.83	0.63	0.21	38.92	500
	FRU50/MF50	15.88			141.70		0.83	0.63	0.21	38.92	500
	FRU50/ STV25/MF25	16.26			60.52	60.25	0.83	0.63	0.21	38.92	500
	STV25/MF75				284.88	70.92	0.83	0.63	0.21	38.92	500

Adapted from Parker, Lopetcharat, & Drake (2018)

FRU50/STV50= 50% fructose/50% stevia, FRU50/MF50= 50% fructose/50% monk fruit, FRU50/STV25/MF25= 50% fructose/25% stevia/25% monk fruit, STV25/MF75= 25% stevia/75% monk fruit.

Table 2. Explanation provided to consumers for sensory terms for temporal check-all-that-apply (TCATA) evaluation of vanilla protein beverages

Attribute	**Explanation**
Cardboard	Aroma/flavor associated with cardboard or brown paper
Sweet Aromatic/Vanilla	Aroma/flavor associated with vanilla, vanillin, or marshmallows
Bitter	Basic taste associated with bitterness
Metallic	Taste associated with metals
Sweet	Basic taste associated with sweetness
Astringency	Tongue-drying sensation

Table 3. Temporal metrics from consumers for vanilla protein beverages with different sweetener systems

		Session 1 Beverage (n=71)				**Session 2 Beverages (n=72)**					
Measure	**Attribute**	**Sucrose**	**Sucralose**	**Stevia**	**Monk Fruit**	**FRU50/ STV50**	**FRU50/ MF50**	**FRU50 STV25/MF25**	**STV25/ MF75**	**Range***	**Range/ Max****
Area Under the Curve	Liking****	1054.72	998.92	922.86	920.24	1089.80	1084.05	1092.97	957.94	172.73	15.8%
	Astringent	35.99	41.89	46.23	52.29	37.83	39.41	41.88	59.22	23.23	39.2%
	Bitter	28.06	18.93	44.24	30.91	34.13	31.93	31.51	46.16	27.23	59.0%
	Cardboard	31.40	18.09	15.70	30.94	26.78	37.08	35.90	41.00	25.30	61.7%
	Metallic	36.10	29.10	56.08	54.95	37.94	31.17	32.43	60.35	31.25	51.8%
	Sweet	59.84	74.85	85.70	75.70	77.97	79.60	88.14	67.09	28.30	32.1%
	Vanilla	75.06	84.91	87.83	76.81	83.04	83.73	81.38	80.08	12.77	14.5%
Peak Citation Proportion	Astringent	0.30	0.34	0.31	0.37	0.27	0.29	0.29	0.42	0.15	36.4%
	Bitter	0.26	0.16	0.40	0.33	0.23	0.22	0.20	0.35	0.25	61.4%
	Cardboard	0.27	0.23	0.18	0.27	0.27	0.28	0.26	0.27	0.10	36.8%
	Metallic	0.28	0.23	0.46	0.44	0.30	0.22	0.24	0.44	0.23	51.3%
	Sweet	0.61	0.64	0.67	0.67	0.57	0.58	0.64	0.59	0.10	15.1%
	Vanilla	0.66	0.74	0.75	0.72	0.64	0.62	0.59	0.58	0.17	22.8%
T_{max}(s)***	Astringent	54	64	61	67	83	51	104	52	53	51.0%
	Bitter	30	22	28	33	31	39	35	37	17	43.6%
	Cardboard	27	28	33	57	30	28	59	61	34	55.7%
	Metallic	22	28	35	30	37	50	25	40	28	56.0%
	Sweet	28	21	30	17	49	47	38	37	32	65.3%
	Vanilla	29	30	30	29	44	49	43	47	20	40.8%

*"Range" refers to the difference between maximum (Max) and minimum values for a given attribute metric.

**Range/Max serves as a standardized value representing variability for a given attribute metric.

***T_{max} = elapsed time in seconds at which peak citation proportion was reported.

****TL was scored on a 9-point scale, whereas TCATA attributes were "scored" according to population proportion that selected an attribute at any given time (citation proportion)

FRU50/STV50= 50% fructose/50% stevia, FRU50/MF50= 50% fructose/50% monk fruit, FRU50/STV25/MF25= 50% fructose/25% stevia/25% monk fruit, STV25/MF75= 25% stevia/75% monk fruit.

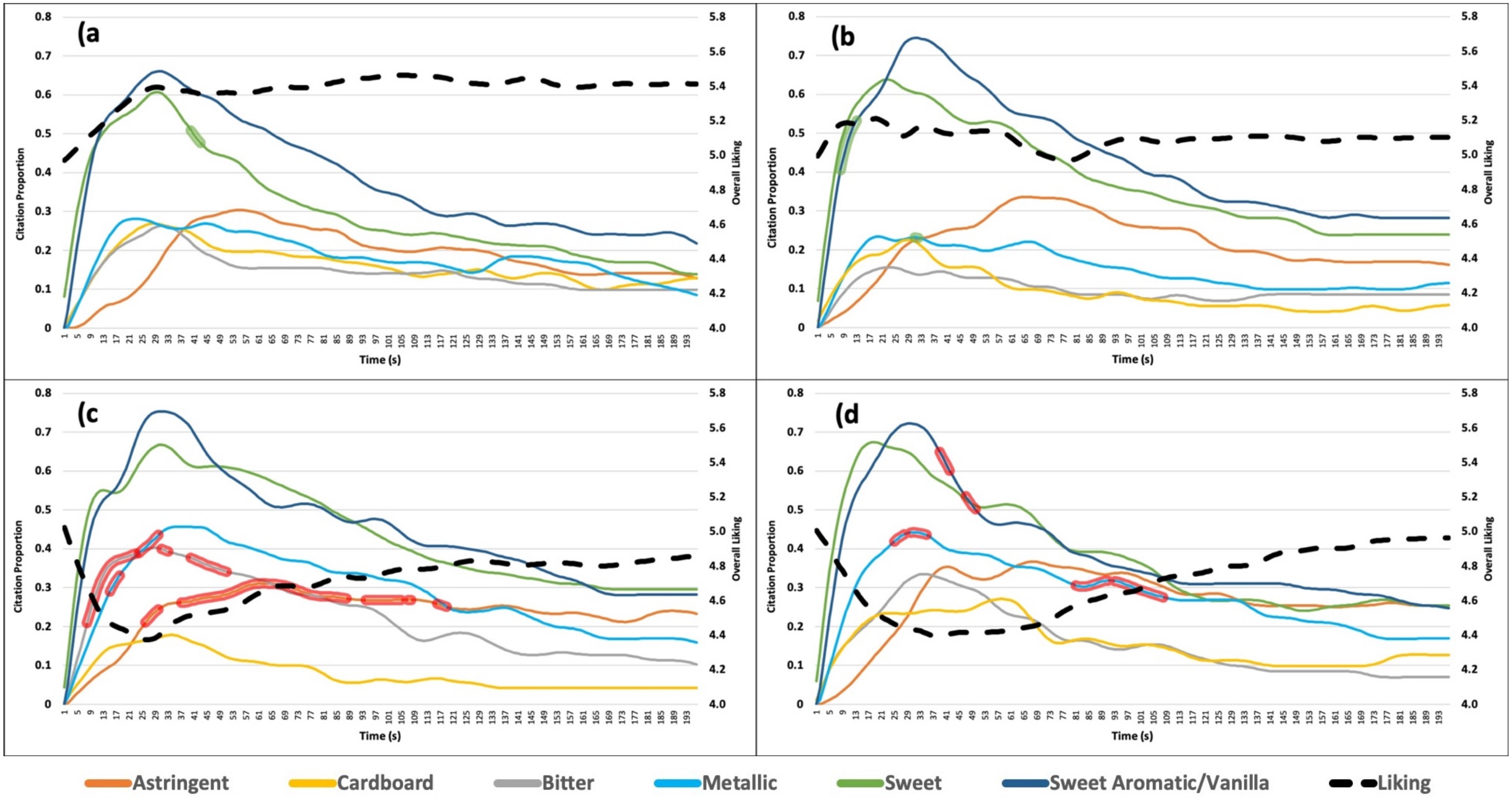

Figure 1. Temporal penalty analysis profiles of protein beverages sweetened individually by a) sucrose, b) sucralose, c) stevia, or d) monk fruit from untrained consumer evaluations (n=71). Overall liking was scored using a 0 to 9 point hedonic scale where 1 = dislike extremely and 9 = like extremely.

*Regions highlighted red indicate significant ($p<0.05$) penalties to liking attributed to perception of a given attribute, and regions highlighted green indicate significant ($p<0.05$) lifts in liking attributed to the perception of a given attribute.

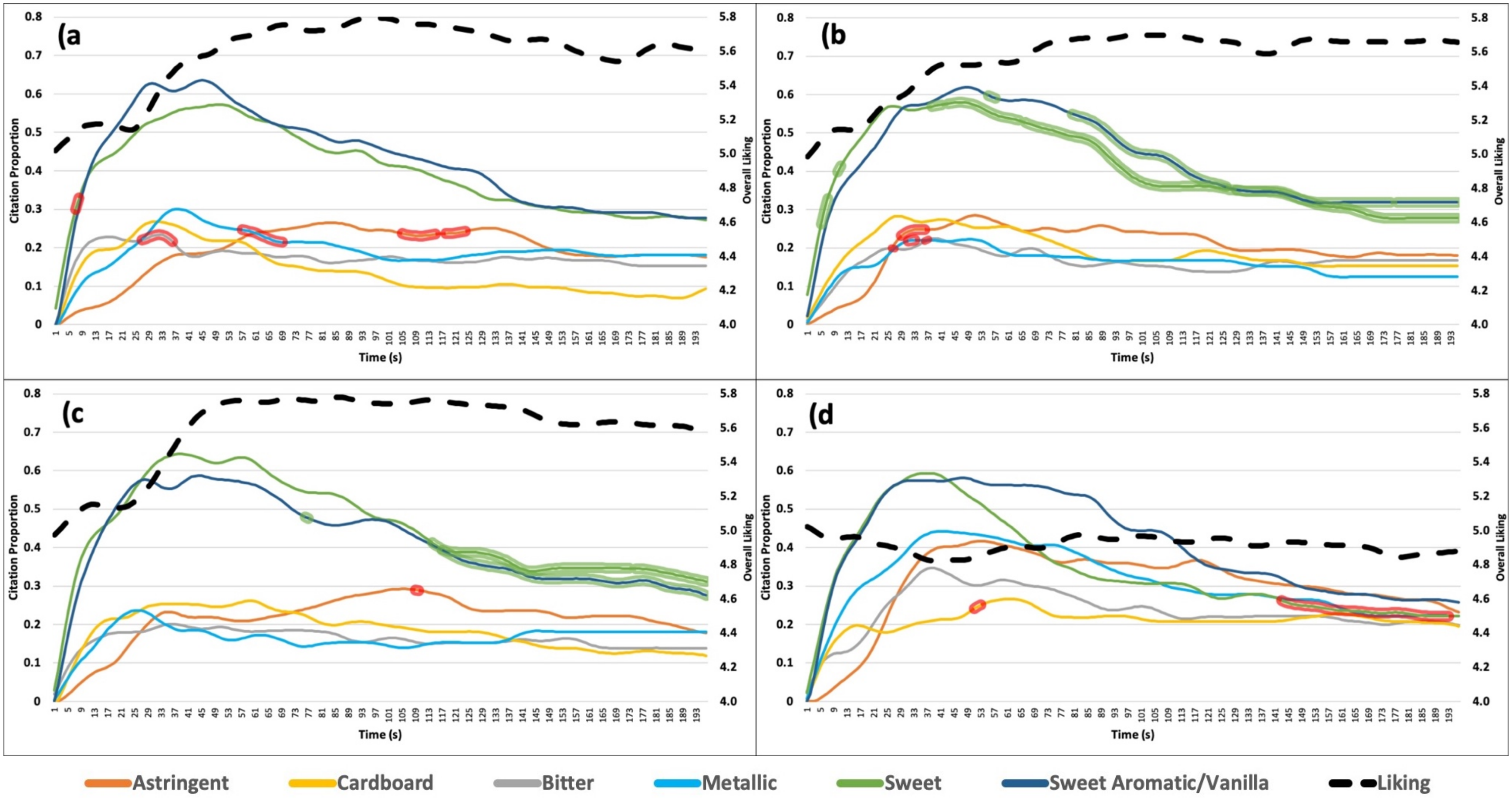

Figure 2. Temporal penalty analysis profiles of protein beverages sweetened by blends of a) 50% fructose/50% stevia, b) 50% fructose/50% monk fruit, c) 50% fructose/25% stevia/25% monk fruit, or d) 25% stevia/75% monk fruit from untrained panelist evaluations (n=72). Overall liking was scored using a 0 to 9 point hedonic scale where 1 = dislike extremely and 9 = like extremely. *Regions highlighted red indicate significant ($p<0.05$) penalties to liking attributed to perception of a given attribute, and regions highlighted green indicate significant ($p<0.05$) lifts in liking attributed to the perception of a given attribute.

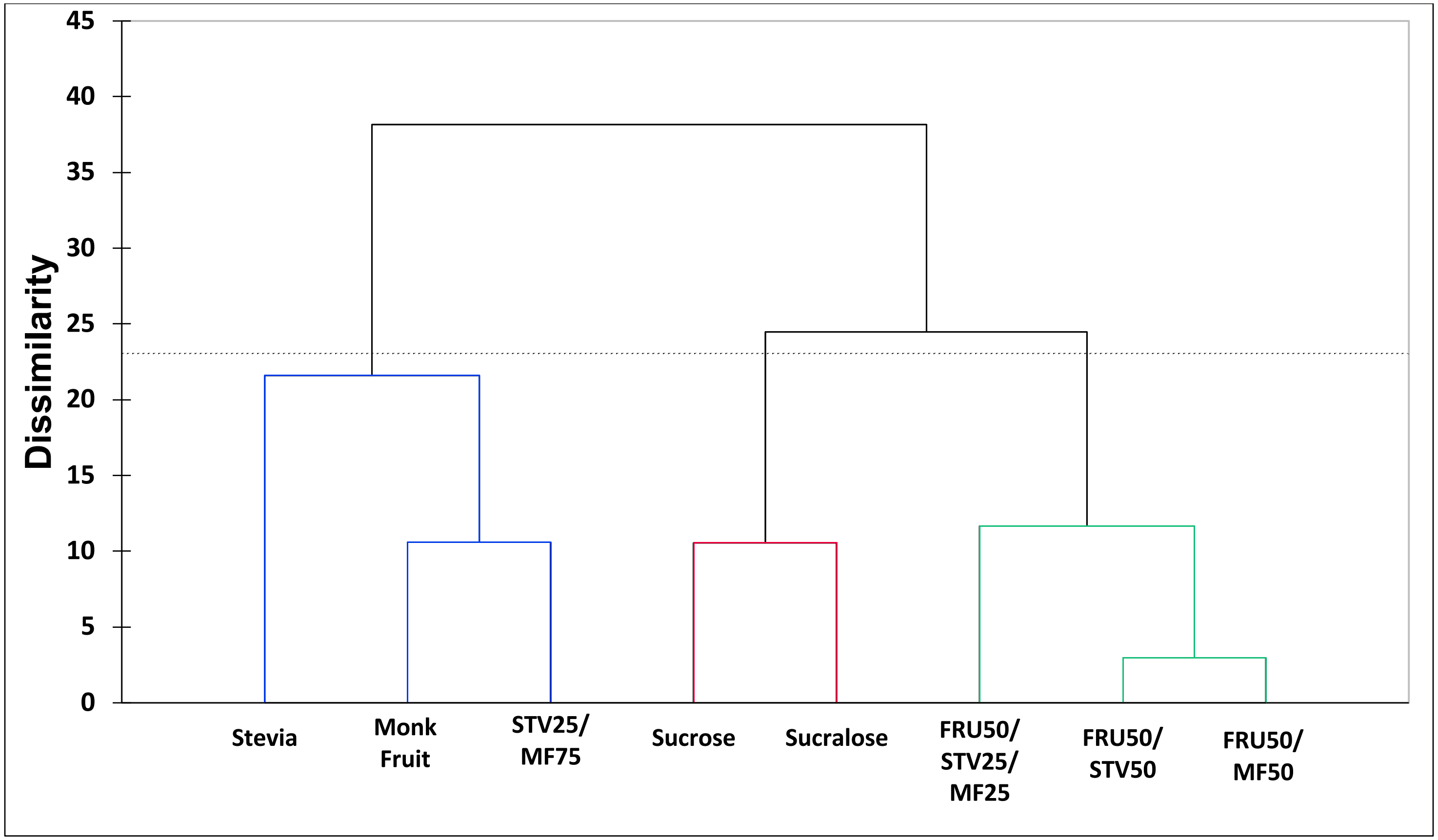

Figure 3. Agglomerative hierarchical clustering (AHC) of vanilla protein beverage formulations with different sweetener systems based upon temporal sensory metrics.

FRU50/STV50= 50% fructose/50% stevia, FRU50/MF50= 50% fructose/50% monk fruit, FRU50/STV25/MF25= 50% fructose/25% stevia/25% monk fruit, STV25/MF75= 25% stevia/75% monk fruit.