ABSTRACT

WAGONER, TY B. Food Structure and its Effects on the Perception of Taste and Texture. (Under the direction of Dr. E. Allen Foegeding).

Food structure can be considered the conformation, connections, and arrangement of molecules in food occurring from molecular to macroscopic length scales. One key property derived from food structure is texture, a sensory percept based on both the underlying food structure and transformations that occur during oral processing. Texture is an important part of the eating experience and has recently been shown to modulate satiation and satiety by extending oral processing time. One hypothesis is that food structure can be leveraged to alter texture, with an overall goal of increasing satiation and satiety while maintaining overall quality. Therefore, the aim of this dissertation was to explore interrelations among food structure, oral processing, and perceived taste and texture. Two types of fluid food structures were investigated – beverages and caramels. Beverages were selected because increased viscosity – desirable for satiety – has historically been associated with decreased taste and aroma perception. Caramels are viscous, adhesive fluids that, unlike beverages, require chewing during oral processing, allowing determination of structural elements associated with adhesion.

The first set of studies focused on texture-taste interactions in beverages. One study evaluated the effects of carboxymethyl cellulose (CMC) concentration relative to its critical overlap concentration (c*) on perceived sweet taste. In CMC solutions, taste suppression only occurred at concentrations well above c*. However, the same concentrations in a dairy-based beverage did not suppress taste, despite higher viscosity. Confocal microscopy revealed microphase separation at CMC levels above 0.8%. The change in perceived sweet taste with increasing viscosity could not be explained due to differences in time intensity, suggesting that delayed temporality does not explain the lack of taste suppression in the dairy system. This indicates that the widely-reported taste suppression in polymer solutions above c* did not apply to beverages with complex microstructures.

A second study established how perceived sweetness intensity of four common sweeteners changed when transitioning from fluid to thick fluid to semisolid food structures. A significant texture-sweetener interaction on iso-sweet taste was observed. The power function slope of non-nutritive sweeteners did not change with food structure; however,

sweetener concentration required for iso-sweet taste and temporal progression of dominant sensory attributes did change. This indicates that sugar reduction strategies are not “one size fits all” in terms of product texture and desired sweetness level.

A third study evaluated the role of mucoadhesion in modifying taste and texture perception in dairy-based beverages. Despite reported differences in mucoadhesion between a random coil polysaccharide and a modified starch, there were few effects on perceived taste and texture. The primary effect was due to hydrocolloid addition increasing viscosity and associated texture terms. However, sensory perception of basic tastes and flavor were not altered. Samples did not exhibit marked differences in the temporal progression of dominant sensory attributes. Results of this study underscore the importance of viscosity in altering texture without a significant effect on taste perception.

For the final two studies, caramels were formulated to investigate how structural elements alter oral processing and texture perception. Hydrocolloid addition significantly increased hardness and reduced adhesiveness. This required more chews, greater jaw movement, and increased jaw muscle activity. This implies a hardness-adhesiveness interaction where jaw movements are altered in response to a combination of both parameters. Adhesion was modeled on viscoelastic parameters determined using creep recovery tests. Notably, components associated with retarded elasticity were associated with increased adhesion. Adhesiveness of samples varying in total protein and fat fell into two zones: one where adhesion was predicted based on surface energy and viscoelasticity, and one based solely on viscoelasticity.

Food Structure and its Effects on the Perception of Taste and Texture

by
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BIOGRAPHY

Ty Wagoner grew up in the small town of Plymouth, IN. Growing up, he was often found in the kitchen helping his parents with such culinary specialties as “busy day meal” and “American spaghetti soup.” After two semesters at Purdue University, he decided to broaden his horizons and pursue a different career path, so he moved 2000 miles across the country to enroll in culinary school at the California School of Culinary Arts in Pasadena, CA. After graduating with an Associate of Science he relocated to Raleigh, NC for an internship at the Second Empire Restaurant and Tavern. He spent years in the food and beverage industry but ultimately desired a more scientific approach. Culinary school teaches the “how” but it often doesn’t answer the “why.” He then decided to enroll at North Carolina State University.

He graduated in 2013 as a class valedictorian receiving a B.S. in Food Science with a Minor in Microbiology. After enjoying an undergraduate research project with Dr. Allen Foegeding, he decided to remain at NCSU to work on a project exploring whey protein thermal stability in beverages as part of his M.S. degree in Food Science, which was conferred in 2015. He opted for the Triple Crown as he remained at NCSU for his Ph.D. He is a third generation Ph.D. following in the footsteps of his grandfather and mother. Upon completion of his Ph.D., Ty will relocate to Berkeley, California to jump headfirst into the world of food biotechnology and gainful employment.

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1. OVERVIEW

As anyone who has just eaten a limp potato chip can attest, texture is a significant part of the eating experience. Textural cues are used to inform the brain of food quality and are also an important driver of liking for consumer preferences, where texture is reported to be a strong driving force in food aversion (Lucas, Prinz, Agrawal, & Bruce, 2004; Nishinari, 2004; Scott & Downey, 2007). Texture can also be leveraged to develop food products with desired characteristics for healthfulness, food quality, and hedonics.

Texture perception is based on food structure. Although sometimes used interchangeably in the literature, structure and texture are not synonymous. Foods have an inherent structure based on the arrangement of molecules in the food. These can be at macroscopic length scales that can be seen (e.g., air bubbles in a foam), or are naked to the human eye (e.g., the protein network in a piece of cheese). Perceived texture, however, is a complex sensory experience that does not refer to a single identifiable quality of food. Whereas taste and flavor are associated with specific receptors, there is no single "texture receptor." Instead, texture arises from physical properties, mechanical stresses, and deformations interacting with a number of different sensory systems (Christensen & Casper, 1987). A significant portion of this process occurs within the oral cavity, due to feedback from thermoreceptors, tactile receptors, and periodontal receptors (Christensen, 1984). Additionally, visual, aural, and extra-oral tactile responses inform a global sensory response. This level of multimodality indicates that the human brain acts as a conduit for texture perception, therefore making it difficult to use mechanical measures of food structure to predict texture.

The past few decades have seen an emerging trend towards increasing the healthfulness of our food supply and altering foods to fit certain types of dietary restrictions (e.g., replacement of gluten for Celiac disease) or to meet the needs of individual consumer groups (e.g., high protein for muscle recovery, softer foods for the elderly). With this comes an interest into how food composition and structure influence the interaction between food

and consumer. Attempts to reduce fat, sugar, and calories in food products often compromise texture in a negative way. For example, the removal of fat tends to make cheese firmer and springier and alters the way the cheese fractures and forms a bolus in the mouth (Gwartney, Foegeding, & Larick, 2002). In addition, there has been increased interest into how food structure and the interaction with food in the mouth (i.e., oral processing) influence overall food intake (Rolls, 2007). The hormonal and neural signals that tell us when and how much to eat begin transmission during oral processing and therefore regulate feelings of fullness and desire to eat (de Graaf & Kok, 2010; Rolls, 2007). Therefore, a comprehensive understanding of how food structure influences texture and oral processing at the macroscale is warranted.

This review of literature is organized into five sections. The first section summarizes molecules commonly used in foods to generate structures, with emphasis on dairy ingredients and polysaccharides. The second section describes how food structure is characterized, providing a basis for subsequent chapters. The next two sections review current knowledge of how food structure alters sweet taste and adhesiveness. The final chapter identifies scientific gaps in the literature and summarizes hypotheses and proposed experiments presented in this dissertation.

2. BUILDING FOOD STRUCTURE

Food ingredients interact at the molecular level to influence macroscopic and functional properties. In designed food systems, the development of food structure often involves the use of protein and/or polysaccharide ingredients (Foegeding, Çakır, & Koç, 2010; Syrbe, Bauer, & Klostermeyer, 1998). It is important to understand the physiochemical aspects of these ingredients because their functionality depends on how they are produced.

2.1. Dairy ingredients

There has been a strong increase in consumer demand for high protein products. Dairy-based foods are in a strong position to meet these needs due to the high levels of essential amino acids and branched-chain amino acids, associations with increased satiety, and delayed gastric emptying (Campbell, Wagoner, & Foegeding, 2017; Ha & Zemel, 2003; Smithers, 2008). From a product development perspective, dairy based ingredients are

popular due to their unique functionality and clean sensory profile (Kelly & Fox, 2016). Functionally, milk is a unique source of protein in that it inherently contains two classes of proteins – caseins and serum (whey) proteins – that are key components in building texture for food products. To understand how dairy ingredients can be used to develop texture in foods, a brief overview of dairy protein ingredient manufacturing is warranted.

All dairy ingredients begin with milk. From the perspective of food structure, bovine milk is a colloidal dispersion of particles spanning multiple size ranges: fat globules in the micron range, casein micelles in the submicron range, serum (or whey) proteins on the order of several nanometers, and remaining small molecules (e.g., lactose, colloidal calcium phosphate) that form true solutions (de Kruif, 1998). These components give milk a total solids content of approximately 12.9%: 3.9% fat, 3.4% protein, 4.8% carbohydrate (primarily lactose) and 0.8% ash, depending on breed, diet, and seasonality (Fox & McSweeney, 2003).

The protein fraction is particularly important in terms of generating structure. Milk proteins were originally categorized into two types on the basis of solubility at pH 4.6: the insoluble caseins and soluble serum or whey proteins. Caseins represent 80% of total milk protein, and include the α_{s1}–casein, α_{s2}–casein, β–casein, and κ–casein subtypes. These proteins differ in calcium sensitivity and presence of thiol groups (Huppertz, 2013). Caseins lack native structure and instead form dispersed micelles aided by the presence of colloidal calcium phosphate and stabilized by hydrophobic and hydrogen bonding among caseins (Thomar & Nicolai, 2016). Casein aggregation in milk is prevented by electrostatic repulsion at milk pH (pH 6.7–6.9) and steric repulsion from κ-caseins (Gebhardt, Takeda, Kulozik, & Doster, 2011; Uversky, Gillespie, & Fink, 2000). However, controlled destabilization of electrostatic and steric forces leads to the generation of food structure in dairy foods such as cheese and yogurt. The remaining 20% of total milk protein is composed of serum, or whey proteins. This collection of proteins includes β–lactoglobulin and α–lactalbumin, with smaller amounts of bovine serum albumin, immunoglobulins, peptides and amino acids. As β–lactoglobulin makes up more than 70% of total serum proteins, it is the primary driver for serum protein functionality (Swaisgood, 1982).

Milk is often consumed as a fluid product or as a starting material for other cultured dairy products. However, the last few decades have seen in increasing use of milk-based fractions as different value-added ingredients. These can be loosely classified as ingredients

1) derived directly from milk, or 2) produced as by-product of cheese-making or acidification processes. A brief overview of fractioned milk ingredients is provided, with particular emphasis on the ingredients relevant to subsequent chapters; additional information can be found in reviews by (Agarwal, Beausire, Patel, & Patel, 2015; Felix da Silva, Ahrné, Ipsen, & Hougaard, 2017; Ikeda, 2015).

The goal of dairy ingredient production is to concentrate the solid components via removal of excess water. For example, the simplest fractionation ingredient is whole milk powder, where water is removed via spray drying to concentrate milk solids without altering the ratio of the individual components. The dried powder contains approximately 36% protein, 52% lactose, 9% fat, with a small amount of ash and residual moisture.

Table 1.1. Common dairy protein membrane filtration techniques and physical properties of the membranes

Membrane	**Pore size (μm)**	**Molecular weight cut-off (kDa)**	**Retentate**	**Permeate**
microfiltration (MF)	0.1 – 10	1000	fat, bacteria, micellular casein	whey proteins, lactose, salts
ultrafiltration (UF)	0.01 – 0.1	10	whey proteins, fat	minerals, lactose, peptides
nanofiltration (NF)	0.001 – 0.01	1	protein, divalent salts, lactose	monovalent salts
reverse osmosis (RO)	< 0.001	0.05	minerals, salts	water

Alternatively, the protein fractions can be concentrated to produce milk protein concentrates (MPC) and isolates (MPI) using membrane separation techniques (Table 1.1). With ultrafiltration, small molecular weight lactose and minerals are removed in the permeate, thereby concentrating proteins in the retentate (Bastian, Collinge, & Ernstrom, 1991). The retentate can then be diluted with water and undergo a second ultrafiltration step – this process is known as diafiltration – to further reduce the non-protein solids prior to

spray drying (Havea, 2006). The resulting ingredients vary in total protein on a dry weight basis from 40–89% and contain the same ratio of caseins to serum proteins as fluid milk (US Dairy Council). Powders that contain greater than 90% protein dry basis are known as milk protein isolates (MPI). Due to the removal of lactose and minerals, MPC is higher in protein and lower in lactose than milk powder (Mulvihill & Ennis, 2003).

Advances in membrane filtration technologies have facilitated the separation of caseins from the serum proteins in fluid milk using microfiltration (Schuck et al., 1994). Up to 95% of serum proteins are removed, with minimal disruption to the casein micellar structure; the resulting ingredient is known as micellar casein concentrate, or MCC (Dalgleish & Corredig, 2012).

Unlike casein-based ingredients, the production of whey protein ingredients historically started as a means of utilizing a waste stream. At approximately 93% water and only 7% solids (of which only 8–10% are protein), a lot of filtration is required to create a protein product from whey streams. However, considering the amount of cheese produced annually in the US, whey has become a major source of valuable dairy protein. Whey generally falls into two production categories: sweet whey and acid whey. Sweet whey is derived from rennet-induced cheese production and has a pH between 6.3–6.6, whereas acid whey is derived from acidified cheeses, such as cottage cheese, and yogurt and has a pH between 4.2–4.4 (Morr & Ha, 1993).

The most widely produced whey protein ingredients are concentrates (WPC) and isolates (WPI) that differ in total amount of protein and lactose. Concentrates are commonly classified by total protein on a dry basis; the two most common WPC products are WPC 35 and WPC 80 (Foegeding, Luck, & Vardhanabhuti, 2011). Both are most commonly produced using ultrafiltration to concentrate protein, with an addition diafiltration step for WPC 80 to further reduce non-protein solids (Kilara, 2008). Whey protein isolate can be made by further concentration of protein solids to greater than 90% via membrane filtration. Ion exchange chromatography can also be used to produce WPI (Huffman & Harper, 1999). Alternatively, the permeate stream from MCC microfiltration can be ultrafiltered to remove excess lactose and minerals and concentrate the serum protein fraction. The resulting spray dried ingredient is known as native whey protein or serum protein isolate to indicate that it is produced from milk and not a derivative of acid or sweet whey.

2.2. Polysaccharides

Dairy ingredients *alone* can be used to generate structure in foods. However, polysaccharides are often added to alter texture, provide mouthfeel, or to enhance gelation, stability, and appearance (Corredig, Sharafbafi, & Kristo, 2011). In dairy products, these commonly include carrageenans, galctomannans, cellulosics, alginates, pectins, or xanthan gum; a summary of these ingredients and their functionalities in beverages is presented in Table 1.2 (Syrbe, Bauer, & Klostermeyer, 1998). Understanding chemical interactions between ingredients in binary mixtures of milk proteins and polysaccharides is critical for controlling structure, functionality, and digestion of the products.

Mixtures of proteins and polysaccharides can produce a macromolecular solution or can lead to associative or segregative interactions, which – depending on the environmental conditions (e.g., pH, ionic strength, temperature) – can either stabilize the system or lead to phase separation and bulk destabilization. Associative interactions arise due to electrostatic attraction between oppositely charged molecules but may also be influenced by hydrogen bonding and hydrophobic interactions to a lesser extent (de Kruif, Weinbreck, & de Vries, 2004; Doublier, Garnier, Renard, & Sanchez, 2000; Turgeon, Schmitt, & Sanchez, 2007). The extent of these interactions determines if large, insoluble complexes (i.e., coacervates) are formed that destabilize the system, or if the ingredients can remain cosoluble under specific environmental conditions.

Segregative interactions, on the other hand, are based on thermodynamics of the system. In binary mixtures of milk proteins and polysaccharides, interpenetration of polysaccharides with large casein micelles is entropically unfavorable. Solvent–polymer interactions are more favorable; therefore, the system separates into two phases (i.e., polymer-enriched and polymer-depleted phases) via a depletion mechanism (de Kruif et al., 2004; Doublier et al., 2000). For example, even at low xanthan gum levels (0.01–0.1% w/w), phase separation occurs in neutral pH dairy beverages after several days (Hemar, Tamehana, Munro, & Singh, 2001). Therefore, in binary mixtures of proteins and polysaccharides, care must be taken to ensure that ingredient usage levels and environmental conditions are controlled to prevent phase separation. These conditions vary based on the type of ingredient; however, a brief overview of polysaccharides relevant to the model systems used within this dissertation is presented.

Table 1.2. Functional properties of hydrocolloids in beverages

Functionalities in beverages	**Mouthfeel enhancement**	**Stabilization of proteins**		**Particle suspension**	**Emulsion stabilization**
Properties of ingredient	**Thickening**	**Protein interaction**		**Network formation**	**Emulsifying properties**
		pH < 4.6	**Neutral pH**		
Alginates	Y (no free calcium)	N	N	Y	N
κ–, ι– carrageenan	Y (at certain temperature)	precipitates	Y	Y	N
λ–carrageenan	Y	precipitates	Y	N	N
Carboxymethyl cellulose	Y	Y	whey separation	N	N
Microcrystalline cellulose	Y	N	N	Y	N
Gellan gum	Y	N	N	Y	N
Guar gum	Y	N	N	N	Y (some grades)
Gum Arabic	Y	N	N	N	Y
Locust bean gum	Y	N	N	N	N
High methoxyl pectin	Y	Y	N	Y	N
Propylene glycol alginate	Y	Y	N	N	Y
Xanthan gum	Y	N	precipitates	N (not xanthan alone)	N

Top rows indicate common functional properties desirable in beverages. Hydrocolloids either have (Y) or do not possess (N) the desired functionality. Table is modified From Paquin et al., 2009.

Carrageenan is commonly used in dairy products due to favorable interactions with casein micelles; these interactions have been leveraged to control the structure of dairy products for decades (Langendorff et al., 1999). Carrageenan is a class of polysaccharides derived from *Crispus rhondus* red seaweed and comprise linear anionic polymers of monomeric 3–linked β–D–galactopyranose, 4–linked α–D–galactopyranose and 3,6–anhydro–α–D–glactopyranose (Campo, Kawano, Silva Jr., & Carvalho, 2009). The three forms of carrageenan found in commercial ingredients (λ–, ι–, and κ–carrageenan) vary in charge density due to the degree of sulfate ester ($–OSO_3^-$) substitution on the carbohydrate backbone. In beverages, low levels of ι– and κ–carrageenan are used to generate a weak gel

network to provide mouthfeel and suspend particulates such as cocoa particles dispersed in chocolate milk. Gelation is a two-step process beginning with a transition from a disordered, coiled state to ordered helical state, followed by aggregation and formation of junction zones that form a gel network. λ–Carrageenan is highly charged and lacks anhydro bridges and therefore does not gel (Rochas, Rinaudo, & Vincendon, 1980).

A second class of polysaccharides commonly used in dairy products is cellulose derivatives, or cellulosics. As the name implies, cellulosics are cellulose-based polysaccharides chemically altered to convert insoluble cellulose into a dispersible and more broadly functional form by modifying hydroxyl groups on the β–1–4 linked anhydroglucose backbone. Cellulosics are characterized by the chemical properties of the substituting group and degree of substitution (i.e., average number of hydroxyl groups substituted per anhydroglucose unit) (Phillips & Williams, 2009). For example, substitution with carboxymethyl esters produces the anionic polysaccharide carboxymethyl cellulose (CMC), whereas hydroxyl propyl ester substitution yields the uncharged but altered polarity hydroxypropylmethyl cellulose (HPMC). These ingredients are non-gelling, water soluble, and can be hydrated at room temperature. Cellulose gum (i.e., CMC) is commonly used in food applications and beverages due to being relatively tasteless and forming clear solutions upon hydration. It is also appropriate for certain targeted applications where its non-caloric, physiologically inert nature is desired (Zhang, 2001). Three common commercial variants are produced based on molecular weight and are referred to as low, medium and high viscosity CMC. The related ingredient microcrystalline cellulose (MCC) or cellulose gel is produced via acid hydrolysis of cellulose to form crystalline aggregate particles. It is often used with CMC in beverages to form a weak three-dimensional network to stabilize particulates against sedimentation (Nsor-Atindana et al., 2017). The degree of polymerization, molecular weight, and percent crystallinity control functionality, but all types require high shear mixing and homogenization for full hydration and dispersion.

3. DEFINING FOOD STRUCTURE

Food scientists interested in the perception of texture have recently begun applying the soft matter physics dogma to food structure. This approach to structure indicates that macroscopic food properties (e.g., taste, flavor, and texture) are not predicted by

physicochemical properties of individual molecules and interactions at the nanoscale *per se* (van der Sman, 2012). Rather, the mesoscale – on the order of microns – predicts macroscopic texture. Our food supply is full of foods that fit this dogma: colloidal dispersions, emulsions, biopolymer gels, fat crystals, and low water foods/high phase volume jammed systems. Thus, understanding how these molecules are structured at this length scale is important for controlling and designing functionality.

In the simplest sense, foods can be categorized on the basis of their physical state: fluids, semi solids, soft solids, and hard solids (Foegeding et al., 2011; van Vliet, van Aken, de Jongh, & Hamer, 2009). Although not all food products neatly fall into this categorization, these states give an indication of food structure, where fluids flow with minimal yield stress, semisolids have fluid-like behavior accompanied by a high yield stress, soft solids fracture into pieces without sound, and hard solids produce pieces and emit sound when fractured (Campbell et al., 2017; van Vliet et al., 2009). Most importantly, these unique food structures are differentiated by their inherent physical properties, the types of sensory perceptions they produce, and how the foods are manipulated during oral processing (Koç, Vinyard, Essick, & Foegeding, 2013). Additional information regarding the interrelations among food structure, perceived texture, and oral processing are provided in Chapter 2; therefore, this section will focus on how food structure is characterized.

3.1. Techniques for characterizing food structure

During consumption, a food is exposed to a range of mechanical deformation processes as it is manipulated in the oral cavity. Therefore, understanding how a material responds to the application of stress is important in characterizing the food. One way to view food structure is through a soft matter physics approach on the basis of molecular length scale. Here the goal is to relate elements of microstructure (at nanometer length scales of molecules themselves) and mesostructure (at micrometer length scales of ordered molecular structures) to macroscopic outcomes such as changes to perceived texture, taste, or eating pattern. Thus, a brief methodological overview of *how* structure is characterized is prudent. Most techniques fall under the umbrella discipline of rheology, or the study of deformation and flow. These tests, which can be empirical or fundamental, involve stress-strain

relationships, to elucidate how a material responds to an applied force (Foegeding, Brown, Drake, & Daubert, 2003).

3.1.1. Viscosity and flow behavior

Rheological characterization of fluids typically involves measuring viscosity. As a material property, viscosity is the resistance of a material to flow (Steffe, 1996), or as a textural term can be thought of as thickness in the mouth. In rotational rheometers, viscosity is determined by measuring the shear stress as a function of a controlled shear strain rate. For Newtonian fluids, viscosity is constant and independent of shear rate. For many food materials, however, viscosity decreases inversely with shear rate; shear-thinning behavior is known as pseudoplasticity.

It has long been a desire of the food industry to use instrumental techniques to develop predictive models for perceived sensory texture. Viscosity has long been the primary property of interest in measuring fluid texture (Szczesniak, 2002). However, predominant wisdom suggests that viscosity *alone* does not fully describe the overall "texture" of a product, although it can provide useful information about some specific texture attributes (Stokes, Boehm, & Baier, 2013). For example, apparent viscosity correlates strongly with visual and oral thickness (Richardson, Morris, Ross-Murphy, Taylor, & Dea, 1989; Stokes et al., 2013). Additionally, viscosity at low shear rates (50 1/s) is reported to correlate with perceived stickiness and mouth coating attributes (He, Hort, & Wolf, 2016). The shear rate of 50 1/s is commonly used as an approximation of in-mouth shear rates in order to compare samples with disparate flow profiles and gives a reasonable prediction of initial viscosity (Stokes et al., 2013). It is important to note, however, that even hydrocolloid solutions with matched viscosity at 50 1/s can vary wildly in viscosity at shear rates above and below 50 1/s depending on the degree of shear thinning. Degree of shear thinning is important in perceived texture and has been reported to correlate strongly with sensory thickness, stickiness, adhesiveness, and mouth coating (Vickers et al., 2015). It has also been reported in inversely correlate with perceived sweetness (He et al., 2016).

Considering the complexity of texture, it is likely that a number of physical properties are contributing to texture perception. For example, creaminess of fluid milk is reported to correlate strongly with both viscosity and lubrication properties, suggesting that increased

viscosity may play a role in higher perceived creaminess (Chojnicka-Paszun, de Jongh, & De Kruif, 2012; Kokini, 1987). In addition to texture, rheological flow behavior plays a role in taste and flavor perception. Early studies reported that increased viscosity leads to a suppression of taste and flavor (Christensen, 1980; Pangborn, Trabue, & Szczesniak, 1973). More recent research points to the concept of molecular structuring, where polymer usage levels exceeding a critical overlap concentration – which is a unique property of a given polysaccharide – were suggested to alter the mixing rate with saliva to suppress flavor and taste (Baines & Morris, 1987; Cook, Hollowood, Linforth, & Taylor, 2002). The fact that there are several sensory modalities stimulated at once also contribute to sensory perception of texture. For example, multimodal integration of viscosity (somatosensory) and other attributes such as aroma and taste perception, have been shown to play a role in texture perception (Rolls, 2004; Stokes et al., 2013). These physical/chemical suppressive mechanisms will be discussed in more depth in section 4.4.

3.1.2. Linear viscoelastic rheological properties

How a material absorbs or dissipates energy is an important consideration that provides insight into the underlying microstructure. In the simplest sense, fluids exhibit viscous behavior by dissipating energy and solids exhibit elastic behavior by recovering energy. However, most complex food systems do not fit neatly into this category and instead fall along a continuum between the two known as viscoelasticity.

The most common method used for measurement of viscoelasticity is small amplitude oscillatory rheology (SAOR). During oscillatory measurements, a sinusoidal stress or strain is applied over the linear viscoelastic range (LVR) – the region where stress and strain are linearly proportional – to the sample as a function of frequency. The total magnitude and time delay of energy transmitted by the sample depend on viscoelasticity. From these tests, a number of useful parameters can be extracted to describe the behavior of the sample: complex modulus (G^*), storage modulus (G'), loss modulus (G''), and phase angle (δ). These parameters give information about food structure that can be related to degree of network formation (Foegeding & Drake, 2007).

Although mechanical spectra give a good indication of inherent structure, they are somewhat limited in terms of predicting perceived texture. For one, SAOR probes the LVR

in order to not damage the sample. The nonlinear viscoelastic region (i.e., outside the LVR) is more relevant to fracture properties of food (Foegeding et al., 2011). The complexity of many food systems also means that SAOR is highly dependent on the time scale of experiments (Stokes et al., 2013). Many texture and mouthfeel sensations arise during oral processing, and SAOR cannot account for the structural transformations that occur in the mouth (Koç et al., 2013). Recent reviews suggest that only certain key elements of food texture – and only at the initial stages of oral processing – can therefore be instrumentally predicted using traditional rheological methods (Stokes et al., 2013).

3.1.3. Creep recovery

Food materials exhibit time dependent behavior, where stress and strain relationships under an applied load are a function of time. To evaluate deformation over longer time scales and to not restrict strain to the LVR, a creep test is used. Creep testing involves the instantaneous application of a stress and plotting strain compliance (J) over time. A purely viscous material instantly dissipates energy whereas a solid material recovers some energy and returns to its original shape. Attributes such as maximum compliance (J_{max}) and retardation time (λ_{ret}) give information about time-dependent behavior. These elements can be fit to viscoelastic models such as the four-element Burger's model, which describes instantaneous elasticity, time-dependent viscoelastic behavior, and long-term viscous flow (Steffe, 1996).

Creep tests are often paired with a recovery test, where the stress is removed and material recovery is monitored. This allows time-dependent viscoelasticity to be evaluated over higher strain rates and longer time scales than SSOR, which are more representative of conditions experienced during mastication (Foegeding et al., 2003). As a result, in some studies creep parameters tended to correlate more strongly with sensory texture than parameters from SAOR (Brown, Foegeding, Daubert, Drake, & Gumpertz, 2003; Wagoner, Luck, & Foegeding, 2016).

As with SAOR, creep parameters provide information about underlying microstructure with the goal of relating to sensory properties. The inverse of J_{max} is often used as an approximation of G′ or firmness. Initial compliance, J_0, related to an undisturbed polymer network; higher J_0 indicates a greater extent of deformation to initiate flow

(Jiménez-Avalos, Ramos-Ramírez, & Salazar-Montoya, 2005). Childs et al. (2007) reported that attributes relating to firmness ($1/J_{max}$) and flow behavior (J_0 and λ_{ret}) correlated strongly with mechanical tack energy during mozzarella cheese shredding (Childs, Daubert, Stefanski, & Foegeding, 2007). This suggests that foods able to flow and better wet surfaces may increase adhesion, which may translate to adhesiveness experienced during oral processing of caramels as well (Wagoner et al., 2016).

3.2. Sensory methodology

The other side of structure characterization takes into account the human interface as a conduit for perceived texture. Whereas rheological techniques probe the inherent structure of a product, sensory analysis gives insight into perceptions of specific textural attributes, which are a product of the underlying food structure and other factors. The inclusion of a human interface gives powerful insight into detection, consumer preferences, and acceptance (Lawless & Heymann, 2010). However, specific texture and mouthfeel attributes are often difficult to define. For example, there is no single texture receptor in the oral cavity; instead, texture percepts are formed from feedback across mechanoreceptors in the hard palate, soft palate, and periodontal membrane, and muscles and tendons associated with mastication (Guinard & Mazzucchelli, 1996).

Quantitation of taste and other sensory attributes can be performed using a number of different sensory analytical tests. These methods are rooted in the discipline of psychophysics, which is the study of physical stimuli and perceptions (Lawless & Heymann, 2010). One of the most powerful sensory tools is descriptive analysis, which comprises a number of different techniques, the most common being Flavor Profile, Quantitative Descriptive Analysis, and Spectrum™ Method. The individual techniques vary in how scales and reference are used, how lexica are established, and how the panel is lead. For example, Spectrum™ is based off a standardized lexicon of attributes decided *a priori* and usage remains constant for similar products over time. Lexica for taste and flavor have been developed with specific references and definitions (Drake, Mcingvale, Gerard, Cadwallader, & Civille, 2001).

4. EFFECTS OF FOOD STRUCTURE ON SWEET TASTE PERCEPTION

One of the goals of understanding food structure at the molecular level is for the design of foods with specific functionality. Therefore, it is important to understand the myriad ways that food structure can alter the way we interact with food. A comprehensive review is outside the scope of this review; therefore, specific outcomes will be the foci of the following sections, beginning with the ways that food structure alters the perception of taste.

4.1. Origin of sweet taste

Considering it is a basic taste (i.e., sweet, salty, sour, bitter and umami) sweetness is anything but basic. From a physiological perspective, the sweet taste percept originates from receptors along oral surfaces and ultimately ends up as an electrochemical response in the brain (Fernstrom et al., 2012). Receptors located within taste buds contain cells that respond to sweet stimuli and send electrical neurotransmissions for cortical processing (Yarmolinsky, Zuker, & Ryba, 2009). These receptors are responsible for stimulus detection and specificity and are Type 1 (T1R) heterodimeric G protein-coupled receptors, specifically T1R2 and T1R3 subunits (Nelson 2001). Each subunit features an extracellular venus-flytrap (VFT) domain, a seven-transmembrane protein domain, and a cysteine rich linking domain (Vigues, Dotson, & Munger, 2009). All sweet tastants bind to this receptor, but differences in sweet quality and intensity are attributed to *how* and *where* tastants bind. For example, dipeptide artificial sweeteners (e.g., aspartame and neotame) bind the T1R2 VFT domain, whereas sucrose, glucose and sucralose bind VFTs on both T1R2 and T1R3 (Nie, Vigues, Hobbs, Conn, & Munger, 2005; Xu et al., 2004).

Despite interest in sweetness mechanisms for over a century, structure-function relationships are still somewhat unknown, and no single model explains all mechanisms of sweet taste (Hayes, 2008). The earliest breakthrough for the molecular interactions that initiate sweet taste was the AH–B model (Shallenberger & Acree, 1967). This model postulates that sweet molecules require two electronegative atoms to act as hydrogen bond donor (AH) and another structural motif to act as hydrogen bond acceptor (B). Although this model does apply to many common sweeteners (including all sweet mono– and disaccharides), it does not explain many high potency sweeteners (Shallenberger, 2009). A modification to this model, incorporates a hydrophobic binding site (Kier, 1972). The most

recent model, called the multipoint attachment theory, denotes eight potential binding sites (Nofre & Tinti, 1996).

4.1.1. Sensory methodology for evaluating taste

Scaling techniques such as magnitude estimation scaling (MES) are often used to determine equivalent sweetness levels (i.e., iso-equivalence concentrations) across different sweeteners (Lawless & Heymann, 2010). With MES, a reference solution is set as a standard and samples are scored relative to the reference solution. The assigned numeric values are arbitrary because only the ratio of perceived intensity relative to the reference is desired. Power functions are then generated based on logarithmic plots of geometric mean sweetness as a function of sweetener concentration. Power functions follow the formula:

$$\log(S) = n \log(I) + \log(k) \qquad \text{Eq. (1)}$$

where S is the magnitude of perceived intensity, k is a proportionality constant, I is perceived intensity given by the stimulus, and n represents exponential growth or decay with concentration (Lawless & Heymann, 2010). The exponent is important because n values less than 1 imply a law of diminishing returns; more and more stimuli must be added for an equal perceived response. Other scaling techniques include category-ratio scales and labeled magnitude scales (or labeled affective magnitude scales) and are derivatives of MES.

Many sensory techniques regard attributes as static responses, but the response is dynamic and occurs over time (Dijksterhuis & Piggott, 2000). It is important to consider the changes that occur during oral processing that alter a food material and, subsequently, taste and texture perception. These transformations include and are not limited to: dilution due to incorporation of saliva, tongue movements that alter mass transfer, temperature changes, movement of air to transfer flavor and alter volatility, breakdown of food structure and release of trapped tastants, and material phase transitions.

Sensory methods such as time intensity (TI) allow attributes to be evaluated over time. This generates a temporal profile or fingerprint of the sample, which provides information such as time to onset, maximum intensity, time to maximum intensity, rate of increase or decrease of intensity, and total duration of intensity (Lawless & Heymann, 2010). Data analysis can be time averaged over all panelists, or individual parameters can be extracted from each individual panelist replication and averaged (McGowan & Lee, 2006).

Time intensity methodology has previously been used to evaluate the progression of sweet taste, development of adhesiveness, flavor release during phase transitions, and texture-taste interactions to name a few (Azevedo, Schmidt, & Bolini, 2015; Bayarri, Rivas, Izquierdo, & Costell, 2007; Hawthornthwaite, Ramjan, & Rosenthal, 2015; Palazzo & Bolini, 2014). This data can also be matched with mathematical models to understand mass transport of tastants in the mouth during oral processing (Le Révérend, Norton, & Bakalis, 2013; Pfeiffer, Boulton, & Noble, 2000).

Time intensity sensory analysis requires a lot of time and only one (and more recently, maybe two) attribute(s) can be evaluated at a given time. Temporal dominance of sensations (TDS) on the other hand allows for the evaluation of multiple attributes at once. Panelists are presented with a selection of attributes and asked to select the most dominant or striking attribute at any given time (Pineau et al., 2009). The selection of attributes is important as only one attribute can be selected at any given time, and only dominant attributes should be offered as a choice to be selected (Ng et al., 2012). The resulting data demonstrate the evolution of several attributes to show product perception pattern over time, providing qualitative insight into sensations that consumers perceive over time (Le Révérend, Hidrio, Fernandes, & Aubry, 2008).

Despite being qualitative in nature, TDS is useful in oral processing studies for evaluating the progression of attributes during mastication. This is especially useful when considering texture-taste interactions as presented in this review, because taste, texture, and flavor attributes can be presented simultaneously, allowing insight into which attributes dominant the sensory experience. This methodology has been useful in evaluating alternative sweeteners for sugar reduction, evolution of bolus attributes during chewing, explaining eating rate and texture perception, and mapping taste, flavor, and texture progression in beverages (Devezeaux de Lavergne, van Delft, van de Velde, van Boekel, & Stieger, 2015; Morais, Pinheiro, Nunes, & Bolini, 2014; Pineau et al., 2009; Young, Cheong, Hedderley, Morgenstern, & James, 2013; Zorn, Alcaire, Vidal, Giménez, & Ares, 2014). Moreover, because it provides qualitative data it is often used to complement traditional sensory profiling (Ng et al., 2012).

A modified form of TDS known as Temporal-Check-All-That-Apply (TCATA) allows multiple attributes to be selected at any given time (Castura, Antúnez, Giménez, &

Ares, 2016). One limitation of TDS is that only one attribute can be selected, so attributes can still be present at relatively strong intensities but just below the most dominant attribute. TCATA overcomes this by allowing multiple attributes to be selected. However, it makes it difficult to discriminant among similar samples as the same types of attributes are likely to be selected. There is also less known about the number of attributes a panelist can evaluate simultaneously (Di Monaco, Su, Masi, & Cavella, 2014).

4.2. Texture-taste interactions in food

Interest in *how* food texture influences taste perception began in the 1960s. Seminal studies by Pangborn et al. and Moscowitz and Arabie were among the first to report taste modification due to the addition of hydrocolloids (Moskowitz & Arabie, 1970; Pangborn et al., 1973). Early studies generally used CMC as a thickening agent and postulated that the results may have been ingredient-specific. However, later studies showed similar effects for increasing concentrations of sodium alginate, xanthan gum, guar gum, carrageenan, and cellulosics (Christensen, 1980; Izutsu, Taneya, Kikuchi, & Sone, 1981; Launay & Pasquet, 1982; Pangborn et al., 1973). This indicates that taste suppression may be a physical principle rather than chemical in nature. Except for the reported increase in sweetness intensity for saccharin in CMC solutions (Pangborn et al., 1973), all studies reported taste suppression as a result of increasing viscosity following a power law function (Izutsu et al., 1981; Moskowitz & Arabie, 1970). These early studies laid down important ground work for texture-taste interactions but are plagued by incomplete rheological data (often only reporting viscosity with no mention of shear rate), narrow shear rate ranges, or not accounting for the contribution of a bulk ingredient like sucrose to viscosity. Moreover, despite a consistent observed effect of viscosity on taste perception, the specific molecular mechanisms at work were not explored.

4.3. Modelling mass transfer of tastants in the oral cavity

Before discussing *how* texture can influence perceived taste, it is first important to present a model for sweet taste perception in response to transient stimuli (Figure 1.1). Based on this mass transfer model, overall perceived taste intensity is proportional to the flux of molecules reaching taste receptors and accounting for differences in taste quality denoted by

factor *b*. The initial starting concentration of stimulus in the oral cavity is known. These molecules must then be released from the food matrix to reach the interfacial layer, and then diffuse through a stagnant salivary layer coating oral surfaces to reach taste receptors.

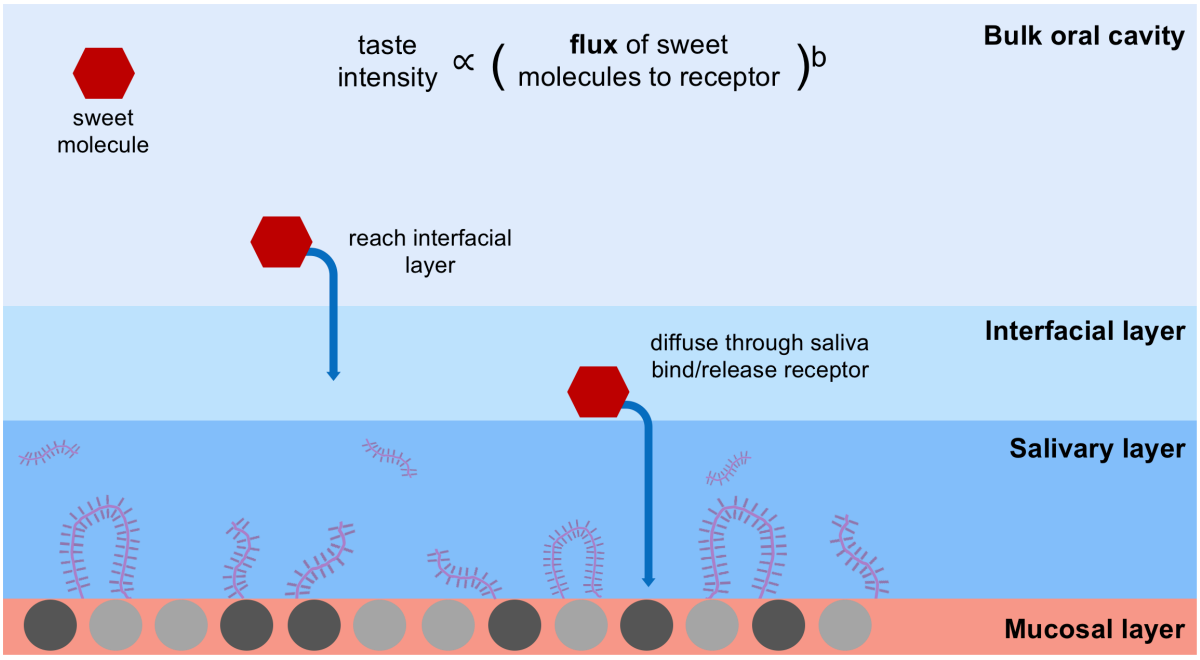

Figure 1.1. Mass transfer model for taste perception. Tastants enter the bulk oral cavity and must reach and pass through an interfacial layer, diffuse through saliva, and bind receptors found within taste buds. Perceived intensity is proportional to the flux of molecules reaching receptors, so structural effects that modify mass transfer should alter taste.

Based on this model, there are three important variables to consider in terms of global taste perception: the bulk concentration of stimulus in the oral cavity, mixing efficiency between the food and saliva, and the proportion of total receptor sites free to bind stimuli (Pfeiffer et al., 2000). The bulk area is assumed to be well-mixed and homogenous, indicating a steady-state equilibrium between the bulk and interfacial regimes (Aubert, Lima, & Le Révérend, 2016). The mucosal salivary layer is assumed to be static; thus, diffusion is the dominant transport phenomenon through the salivary layer (Le Révérend et al., 2013). Additionally, the protrusion of papillae creates a fluid boundary layer where diffusion drives mass transport and thus low shear viscosity is important (Aubert et al., 2016).

Diffusion for sweet and salty stimuli behaves in accordance with Fick's Law, where molal flux is driven by stimulus molecular weight and the concentration gradient spanning the salivary layer (Kokini, Bistany, Poole, & Stier, 1982; Le Révérend et al., 2013; Pfeiffer et al., 2000). Binding kinetics between tastant and receptor are dependent on the number of free binding sites and the concentration gradient across the salivary layer, with an upper theoretical limit associated with receptor saturation (Pfeiffer et al., 2000). This binding behavior is based on apparent binding affinity, which is the substrate concentration at which half of receptor sites are bound (Pfeiffer et al., 2000; Shamil, Birch, Jackson, & Meek, 1988).

In terms of this taste perception model, one could argue that the two most important parameters of a sample are viscosity at low shear rates (as Fickian diffusion through the salivary layer would be governed by low-shear viscosity within the stagnant region) and how well a material mixes with saliva in the bulk region (Le Révérend et al., 2013). The assumption that low shear viscosity drives taste perception has been corroborated by recent studies, where taste perception of hydrocolloid-thickened fluids correlated strongly with viscosity below 50 1/s (Aubert et al., 2016; He et al., 2016; Koliandris et al., 2010). It should be noted that this model is based on fluids but would also apply to solids foods accounting for an additional step to release tastant molecules from a food matrix. This likely explains why solid foods that express more serum or fracture into a greater number of particles during oral processing (i.e., greater surface area) are rated as sweeter (Bayarri et al., 2007; Mosca, van de Velde, Bult, van Boekel, & Stieger, 2015). It is also important to note that mathematical models do not take composition of the salivary layer into account.

4.4. Molecular mechanisms influencing taste perception

4.4.1. Diffusion

Based on this model, there are multiple ways in which food microstructure can alter taste perception. One mechanism suggests altered diffusion of tastants on the basis of increasing viscosity. Assuming the bulk region is infinitely well mixed, mass transfer happens nearly instantaneously, which agrees with the reported sweet taste response time of 50 ms (Shallenberger, 1997). This would suggest diffusion is not the limiting factor in moving from the bulk region to a localized taste bud. However, owing to the static mucosal layer (again, where low shear rates are likely to occur), mass transfer is dominated by Fickian

diffusion. This implies an effect of molecular weight and hydrodynamic size on taste perception due to their effect on tastant diffusivity and may explain differences in temporal sweetness of large molecular weight sweeteners. The effect of diffusivity has been modeled and fit to time intensity data of sweet taste for sucrose, aspartame, brazzein, monellun, and thaumatin (Pfeiffer et al., 2000). Diffusivities ranged from 11.0 – 64.5 m^2/s and molecular weights ranged from 342 Da (sucrose) to 22,209 Da (thaumatin). Accounting for these differences, the model strongly correlated with sensory data (Pfeiffer et al., 2000).

Although the effect of viscosity on diffusion has long been implied in taste suppression, Kokini et al. were the first to quantify diffusion rates (Kokini et al., 1982). In the study, diffusion coefficients of sucrose and fructose were measured in a two-compartment diaphragm cell with a semipermeable membrane by determining the change in refractive index over time. Increased amounts of tomato solids – primarily pectin and cellulose – correlated with reduced diffusion rates and lower perceived sweet taste intensity. More recently, Le Révérend et al. (2013) derived a mathematical model for taste perception vis-à-vis oral processing and fit it to published sensory data. The authors summarized by suggesting that increasing viscosity appeared to decrease mass transfer through the salivary layer. Reduced diffusion coefficients were also measured by (Han et al., 2014) for increasing concentrations of CMC, but not for sodium alginate. A summary of these studies indicates a probable role of diffusion in taste perception but there are likely other contributors to such marked differences in sweet taste that have been reported (Cook, Hollowood, Linforth, & Taylor, 2002). As a result, it is still unclear how much of a practical difference altering diffusion makes, or if it even manifests as a perceptual difference.

4.4.2. Ingredient structuring

Although many early studies from the 1970's observed taste suppression in fluids with high concentrations of hydrocolloids, Baines and Morris were the first to propose a mechanism related to the microstructure of the system (Baines & Morris, 1987). At low usage levels, guar gum had no effect on taste perception; however, at high enough concentrations sweet taste decreased. This inflection point corresponded to the critical overlap concentration, c*. At concentrations below c*, hydrocolloids are dispersed and non-interacting. However, at concentrations above c* the hydrocolloid chains overlap in

hydrodynamic volume and form a transient network associated with a reduction in molecular movement and a marked increase in viscosity. The authors hypothesized that in this overlapping system, inefficient mixing between solution and saliva results in perceptual changes (Baines & Morris, 1987).

This mechanism of taste suppression above c* is strongly validated when examining the bulk of the literature in this area (Table 1.3). This phenomenon has been observed with both nutritive and non-nutritive sweeteners for a number of polymers, including cellulosics guar gum, and carrageenans (Baines & Morris, 1987; Cook et al., 2002; Cook, Hollowood, Linforth, & Taylor, 2003; Han et al., 2014; Hollowood, Linforth, & Taylor, 2002). Additionally, different hydrocolloids align to a master curve when sensory data is plotted as a function of the Kokini oral shear stress (Cook et al., 2003).

Table 1.3: Effects of polysaccharide concentration on sweet taste relative to the critical overlap concentration (c*).

Ingredients	**Effect below c***	**Effect above c***	**Reference**
λ–CG	None	None	Cook et al., 2002
λ–CG	None	Reduction	Cook et al., 2003
CMC	None	Reduction	Han et al., 2014
Guar gum	None	Reduction	Baines & Morris, 1987
Guar gum	None	Reduction	Cook et al., 2002
Guar gum	None	Reduction	Cook et al., 2003
HPMC	None	Reduction	Han et al., 2014
HPMC	Slight reduction	Slight reduction	Hollowood et al., 2002
HPMC	None	Reduction	Cook et al., 2002
HPMC	None	Reduction	Cook et al., 2003

CMC, carboxymethyl cellulose; HPMC, hydroxypropylmethyl cellulose; CG, carrageenan

Interestingly, inefficient mixing of polymers above c* may also explain why taste suppression is not as severe in starch-thickened foods at similar viscosities (Hill, Mitchell, &

Sherman, 1995). Structurally, starch granules represent particulate systems with significantly less overlapping volume in which to trap molecules. A comparison of wheat, waxy maize, modified waxy maize, and hydroxyl propyl methyl cellulose (HPMC) at similar viscosities showed that both flavor and salty taste were weakest in the polymeric HPMC system, and strongest in the wheat and modified maize systems where granular integrity was maintained (Ferry et al., 2006). Confocal microscopy indicated that waxy maize exhibited partially disintegrated granules (i.e., a combination of polymeric and particulate systems) and taste intensity fell in the middle. However, it should be noted that the presence of salivary α–amylase can reduce the viscosity of starch pastes in a few seconds but would have no effect on HPMC (Ferry, Hort, Mitchell, Lagarrigue, & Pamies, 2004).

The idea that polymer concentrations above c* result in inefficient mixing of saliva and sample would imply that pockets of tastants are physically obstructed from reaching a receptor. As a fluid is manipulated during oral processing, these pockets would be broken up and diluted with saliva. In the case of two iso-sweet polymers solutions – one above and one below c* for a given polymer – the same number of sweet stimuli would be available. However, above c* the stimuli would be delayed as a result of inefficient mixing – thus, an altered temporal pattern of sweetness would be expected. As of now, no studies have evaluated the temporal pattern of sweet taste at concentrations above and below the c*.

4.4.3. Binding

The original hypothesized mechanism of taste suppression by hydrocolloids was attributed to molecular interactions with tastant molecules (Pangborn et al., 1973). Covalent bond formation, hydrophobic interactions, and electrostatic interactions between hydrocolloids and *flavor* molecules have been documented (Roberts, Elmore, Langley, & Bakker, 1996; Secouard, Malhiac, Grisel, & Decroix, 2003). However, the role of binding in *taste* perception remains unclear.

The concept of molecular binding could imply directional interactions such as those documented for aroma compounds. For example, at pH 5–7 aspartame adopts a zwitterionic form that could electrostatically interact with charged polysaccharides such as λ–carrageenan (Cook et al., 2002). This has been proposed but not experimentally shown. Alternatively, the addition of hydrocolloids could indirectly alter binding equilibria between tastant and taste

receptors. Increasing concentrations of sodium alginate and CMC have been reported to decrease the association constant between aspartame and a fullerenol model receptor (Han et al., 2014). This was associated with a loss of water mobility at concentrations above c*.

4.4.4. Cross modal interactions

Perception of taste and texture is the interaction of olfactory, gustatory, and somatosensory tactile sensations (e.g., thickness, creaminess, mouthcoating, adhesion), which underscores the role of cognition in total taste perception. Combinations of these signals enter the orbitofrontal cortex for processing and integration into a global response (Calvert, Brammer, & Iversen, 1998). This makes it particularly challenging to study the effects of texture on taste or flavor perception – even in simple polymer and water solutions – because modifying viscosity may also modify flavor release and mouthfeel attributes (Cook et al., 2003; Ferry et al., 2006). For example, the activity of orbitofrontal cortex neurons is altered by increasing viscosity or by transitioning from liquid to semisolid foods (Rolls, 2005). Interestingly, this texture-taste interaction appears to be non-reciprocal, where increasing viscosity decreases sweet taste but adjusting sweet taste doesn't modify perceived texture (Tournier et al., 2009).

Taste-aroma (i.e., flavor) interactions have been well established at both subthreshold and suprathreshold levels. Increasing volatile aroma compounds in sweet model systems was reported to increase perceived sweetness (Clark & Lawless, 1994; Hort & Hollowood, 2004). This interaction appears to be reciprocal, where increased sweet tastant concentration was reported to increase fruity aroma intensity (Hollowood et al., 2002; Hort & Hollowood, 2004; Pfeiffer et al., 2000). Accordingly, increasing viscosity by addition of hydrocolloids could contribute additional taste and aromas that modify perceived taste. Conversely, the hydrocolloids may bind volatile compounds without altering mass transfer of tastant to receptor and still be perceived as less sweet. This also suggests a strong cognitive component, as more viscous samples have been reported to have less intense flavor despite the measured concentration of volatiles in the nasal cavity being unchanged (Cook, Hollowood, Linforth, & Taylor, 2005; Hollowood et al., 2002).

Compositional differences can also alter perception. For example, not only is a fat an inherent source of flavor but it also modifies taste and flavor perception. In oil in water

emulsions, the effect of fat level on perceived aroma and taste was more pronounced than the effect of increasing viscosity with guar gum (Malone, Appelqvist, & Norton, 2003). In complex, multicomponent systems, the texture-taste effect could be related to binding of aroma molecules. Binding of volatile compounds by proteins has been well established, where the presence of casein or whey proteins has been reported to decrease aroma intensity of benzaldehyde and D–limonene (Hansen & Heinis, 1992). Viscosity also appears to play a role, where solutions thickened with CMC or guar gum reduced intensity of α-pinene and ethyl 2–methylbutyrate compared to water (Roberts et al., 1996).

5. EFFECTS OF FOOD STRUCTURE ON ADHESIVENESS

One goal for understanding food structure is to understand how to alter or control textural attributes. The effects of modifying microstructure on taste and texture perception have been reviewed extensively (Foegeding, Çakır, & Koç, 2010; Kuo & Lee, 2014; Stieger & van de Velde, 2013). However, adhesiveness is one particular textural attribute of the eating experience that is often overlooked. Despite being one of the original five attributes tested with the texture profile analysis (TPA) method, the mechanisms that ultimately control and alter adhesion are not well understood (Friedman, Whitney, & Szczesniak, 1963). Adhesion is typically viewed as an undesirable attribute, where sticking to processing surfaces, packages, or oral surfaces during chewing is undesirable. This is the primary reason consumers avoid certain types of foods, especially with elderly consumers that may have decreased chewing efficiency or artificial teeth (Peleg, 1993). However, adhesiveness is an important attribute that is thought to inform the brain about the readiness and safety of a food to be swallowed (Loret et al., 2011). It may also be an expected or desired attribute for certain types of food products such as caramels and other confections.

5.1. Adhesiveness during oral processing

Sensory perception of adhesiveness can be viewed in two ways: sticking to teeth (which is often correlated with the force required to remove from these surfaces) and the idea of mucoadhesion or a lingering film on oral surfaces. Total adhesive strength is governed by physical properties of the material and contacting surface (e.g., rheology, glass transition temperature, ingredient hydrophobicity, and surface chemistry) and external conditions such

as applied pressure, contact time, temperature, contact surface area, and presence of lubricant (Adhikari, Howes, Bhandari, & Truong, 2001; Bhandari & Howes, 2005; Foley & Chu, 1986; Saunders, Hamann, & Lineback, 1992). The most common method for measuring adhesiveness is via mechanical evaluations in compression. This is done empirically, as part of compression-based tests such as instrumental texture profile analysis, or under controlled conditions where a material is compressed to a given distance or at a given force, held for a given amount of time, and then released. Mechanical adhesion can then be quantified as the total area under the debonding curve (Kim et al., 2009), the peak force upon decompression (Wagoner et al., 2016), or the slope of the negative debonding curve (Sai-Manohar & Haridas-Rao, 1997).

One goal is to combine mechanical tests with sensory adhesiveness to understand how material properties influence overall perceived stickiness. However, mechanical measures often do not incorporate saliva or account for material transformations that occur during oral processing; as a result, they often poorly correlate with sensory data (Kim et al., 2009; Pascua, Koç, & Foegeding, 2013). Therefore, sensory techniques that provide insight by accounting for material transformations, phase transitions, and the incorporation of saliva over time are more useful. Temporal sensory techniques such as TDS, TCATA, and TI are useful if adhesion is a dominant attribute. Time intensity of stickiness has been used to differentiate nut pastes with various degrees of added oil (Hawthornthwaite et al., 2015). The temporal aggregate curves were similar, but maximum stickiness intensity and total duration decreased with oil up to 10% added oil. For low moisture foods such as nut pastes and dry biscuits, TDS revealed that stickiness began to develop towards the end of the oral process, likely due to the incorporation of saliva during oral processing (Hawthornthwaite et al., 2015; Young et al., 2013).

Typically, relationships among food structure and adhesiveness have been evaluated in model foods such as biopolymer gels and process cheese, or in products where adhesion is a dominant attribute. Adhesiveness can be modulated by altering the formulation, such as increasing total fat content or by the addition of biopolymers (Çakir et al., 2012; Childs et al., 2007; Mendenhall & Hartel, 2016; Steiner, Foegeding, & Drake, 2003; Wagoner et al., 2016). An increase in adhesiveness has a profound effect on oral processing patterns. It has been associated with increased total duration of oral processing, greater number of chewing

cycles, slower chewing rate, and greater jaw range of motion (Çakir et al., 2012; Kohyama et al., 2005; Wagoner et al., 2016). Ultimately, these attributes – in addition to food viscosity and bite size – are critical components associated with food intake and satiety (Hogenkamp & Schiöth, 2013).

5.2. Mucoadhesion

Related to the *macroscopic* idea of adhesion or stickiness is the idea of mucoadhesion, or adhesion of molecules to mucosal surfaces in the oral cavity. This concept has long been leveraged by the pharmaceutical industry to enhance drug delivery by ensuring contact of a compound with mucosal surfaces for a longer time (Cook, Bull, Methven, Parker, & Khutoryanskiy, 2017; Smart, 2005). It has only been recently that this concept has arisen in the food science literature as an important attribute for sensory texture. Many of the same polysaccharides used as mucoadhesives in pharmaceuticals are also used in food formulations. Additionally, many food components inherently exhibit mucoadhesive properties, such as milk proteins and polyphenols. Not only do these ingredients have the capacity to alter organoleptic properties of foods, but they also have the potential to be leveraged in order to control specific attributes.

Understanding mucoadhesion begins with understanding the tissues that line all oral surfaces. Generally, three main types exist: masticatory mucosa covering the hard palate, non-keratinized lining mucosa, and specialized mucosa primarily found on the dorsal side of the tongue (Cook et al., 2017). In the mouth, mucosal surfaces are covered by a 1–100 μm layer of saliva (Smart, 2005). Saliva is approximately 99% water, with the solid content composed of various salts, proteins, and phospholipids. The protein fraction is composed of amylase, proline rich proteins, mucins, cystatins, histatins, and statherins (Carpenter, 2013). However, mucins are the main component responsible for the viscoelasticity, surface active behavior, and unique physical properties of saliva, and are therefore the most important component in mucoadhesion (Bansil & Turner, 2006).

The unique functionality of mucins is attributed to structural conformations. Mucins contain a protein core with a high degree of glycosylation, often making up more than 80% of the total weight. These oligosaccharide side chains are covalently bound to regions with high amounts of proline, threonine, and serine (Thomsson et al., 2002). The chains terminate

with sialic acid or sulfonic acid residues, giving a net negative charge under physiological conditions (Gandhi & Robinson, 1994). Hydrogen bonding among sugars and hydrophobic interactions among nonpolar groups both give mucins a tendency to aggregate into suprastructures, giving a large distribution of sizes ranging from 0.5 to 20 MDa (Bansil & Turner, 2006). Mucins exist both in saliva and as transmembrane proteins covering epithelial cells on the oral mucosa. Saliva primarily contains a high molecular weight MUC5B fraction, whereas the mucosa is covered by a mix of MUC5B and low molecular weight MUC7 fractions (Schipper, Silletti, & Vingerhoeds, 2007).

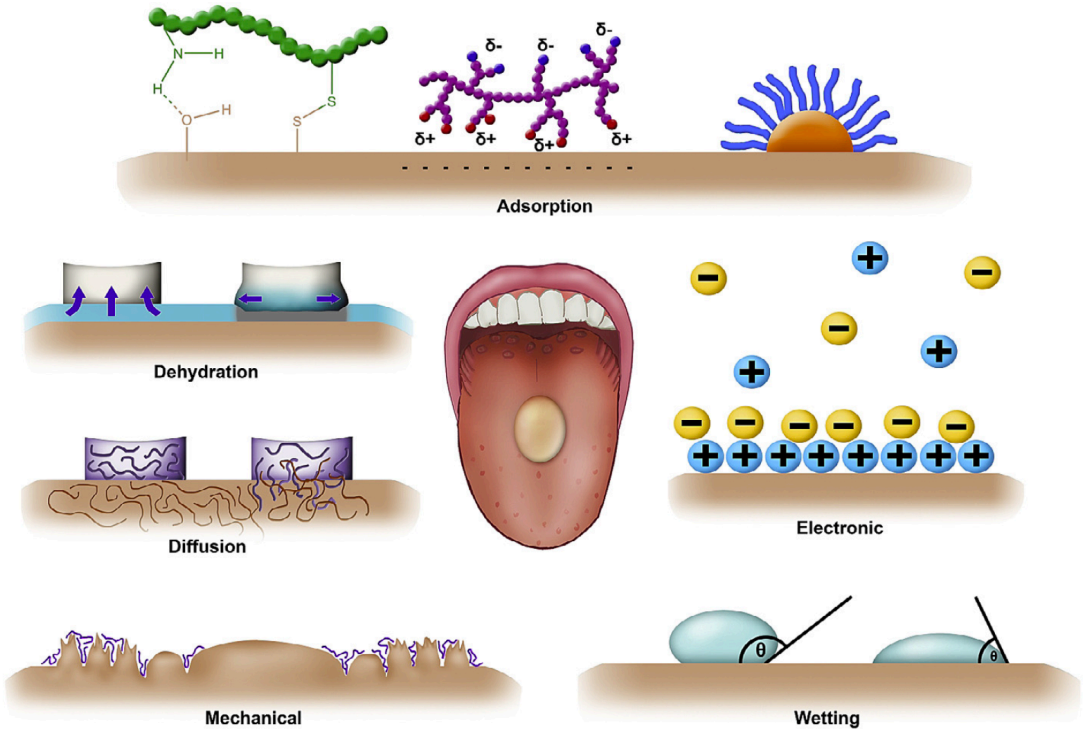

Figure 1.2. Theorized mechanisms by which mucoadhesive polymers adhere to mucosal surfaces. Adsorption includes hydrogen bonding and disulfide interactions, dipole interactions with negatively charged mucins, or hydrophobic portions of colloids interacting with mucins. Additional mechanisms include dehydration, diffusion, mechanical interactions, electronic interactions, or wetting phenomena. From Cook et al., 2017 with permission by Elsevier Ltd.

Mucoadhesion is based on the principle that compounds in food interact with mucins and mucosal surfaces. Considering the complexity of mucin structure and the wide range of molecules that are mucoadhesive, there are six fundamental theories that describe these interactions. A brief summary is presented here; more detailed information about these theories can be found in a number of reviews (Andrews, Laverty, & Jones, 2009; Khutoryanskiy, 2011; Smart, 2005). Figure 1.2 depicts the six theories of mucoadhesion: adsorption, dehydration, diffusion, electronic, mechanical, and wetting. Adsorption is based on general non-covalent interactions (e.g., van der Waals, hydrogen bonding & hydrophobic interactions) between mucins and polysaccharides, whereas electronic theory describes electrostatic interactions. Dehydration theory is predicated on a moisture disparity between two components, where movement of water from mucus into the second material enhances adhesion. This is most common to solid, water-absorbing materials (Jabbari, Wisniewski, & A. Peppas, 1993). Entanglement and interpenetration of compounds comprises diffusion theory. Mechanical and wetting theories are related and describe contact angles and spreading ability, respectively.

5.2.1. Mucoadhesion quantitation methodology

Quantitation of mucoadhesion usually falls into *in vitro* (or *ex vivo*) methods or by evaluating physical properties as an indication of adhesive strength (Cook et al., 2017). One method is to measure the detachment force or debonding curve between an adhesive and mucosal surface. Other groups have developed flow-through retention experiments where an irrigation media is flowed over an adhesive coating mucosal tissue. Quantitation involves fluorescent imaging of the mucosal tissue and colorimetric assays of the eluent over time (Cave, Cook, Connon, & Khutoryanskiy, 2012). Tribology can also be used to assess the friction between adhesive and mucosal analogs in order to relate to sensory properties (Dresselhuis, Aken, Hoog, & Stuart, 2008).

Physical methods tend to use solutions of mucin particles rather than mucosal tissue, with the assumption that interactions between mucins and mucoadhesive ingredients will alter physical properties of the solution. This can be measured as a change in solution viscosity, viscoelasticity, or optical density (Cook et al., 2017). Several groups have used a combination of dynamic light scattering and electrophoretic mobility to elucidate the change

in particle size and zeta-potential, respectively (Takeuchi et al., 2005). Other techniques involve isothermal titration calorimetry and differential scanning calorimetry to evaluate binding kinetics, and circular dichroism to study conformational changes as a result of binding (Cook et al., 2017).

5.3. Mucoadhesion and food

Despite the recognition of mucoadhesion by pharma, it is still a rather novel concept vis-à-vis food consumption. The earliest mention in food science literature was a hypothesis that reported differences in taste and flavor among three polysaccharide thickeners may have been due to differences in mucoadhesion (Mälkki, Heiniö, & Autio, 1993). Mucoadhesion is still rarely assessed or directly mentioned yet is thought to be an ancillary contribution to mouthfeel sensations such as astringency and mouth-coating (Cook et al., 2017). Research on mucoadhesion typically falls into one of three categories: relationship with astringency and mouthfeel attributes, role in altering hydration and lubrication in the mouth, or altering mass transfer of molecules.

Astringency and mouth drying are typically viewed as negative attributes, although astringency of polyphenols and tannins in wine can be desired. The mechanisms that contribute to astringency are not yet fully elucidated but are thought to be due to interactions among food components and salivary proteins that reduce lubrication over the oral surfaces – hence the drying sensation (Gibbins & Carpenter, 2013; Lee & Vickers, 2012; Rossetti, Yakubov, Stokes, Williamson, & Fuller, 2008). In food systems, astringent compounds include those commonly found in wine, tea, fruits, or dairy products. Additionally, many of the same polysaccharides that are used to control delivery of drugs through mucosal surfaces by the pharmaceutical industry exhibit this quality (Smart, 2005). Notably, the cationic polymer chitosan is considered a strong mucoadhesive and also exhibits strong astringency when adsorbed to oral surfaces (Luck, Varum, & Foegeding, 2015). Processing conditions can also alter astringency; increased heating time of whey protein beverages lead to increased particle size and higher perceived mouth drying (Bull et al., 2017). It has also been proposed that mucoadhesion could be leveraged to control astringency. One study showed that the presence of CMC (a strong mucoadhesive) reduced astringency of polyphenolic compounds, which the authors hypothesized was due to competitive binding with salivary mucins

(Troszynska et al., 2010). Additionally, a recent study reported that retention of sodium ions on a tongue increased with increasing CMC concentration, which had the effect of altering temporal taste perception and diminishing maximum perceived taste intensity (Cook, Woods, Methven, Parker, & Khutoryanskiy, 2018).

The ability of compounds in food to bind to mucins on oral surfaces and in saliva will have an effect on lubrication in the mouth. Spreading over oral surfaces is important to sensory attributes such as smoothness, creaminess or other attributes that may be desired and are particularly difficult to replace in low fat food formulations. Creaminess, for example, is partly due to the flavor and lubricating quality of fats, but also the frictional forces among saliva, food matrix, and oral surfaces (Jervis, Gerard, Drake, Lopetcharat, & Drake, 2014; Selway & Stokes, 2014). It has been reported that emulsified fats are more likely to adhere to oral surfaces and coalesce with salivary proteins, enhancing lubrication and sensory perceptions of creamy and fatty mouth feel (Dresselhuis et al., 2008). Controlled mucoadhesion may be a method of enhancing lubrication on oral surfaces, allowing better design foods with controlled mouth attributes (Malone et al., 2003; Stokes, Macakova, Chojnicka-Paszun, de Kruif, & de Jongh, 2011).

Last is the nascent idea of controlled mucoadhesion for designed functionality. Judicious selection (or chemical modification) of mucoadhesive ingredients could be a way to control release of tastant or flavor molecules or to alter the temporal progression of these attributes. For example, volatile compounds can bind either directly to oral mucosa, or to mucoadhesives adsorbed onto mucosa (Malone et al., 2003). This can influence aroma persistence and perceived taste and flavor over time. At this point, the primary challenge is separating the contributions of viscosity, adhesion between food components and oral surfaces, and texture-taste interactions to overall sensory perceptions.

6. SCIENTIFIC GAP ANALYSIS, HYPOTHESES, AND OUTLOOK

Relationships between mechanical or physical properties and perceived texture during oral processing have been explored but are still not well understood. For example, adhesiveness and cohesiveness are texture attributes that have received little attention in the literature, and there is little known about the properties of a food and the transformations that occur in the mouth that contribute to perceived adhesiveness. Additionally, oral processing

studies commonly use model food gels that can be fine-tuned such as process cheese or biopolymer gels. Therefore, the effects of compositional differences on oral processing behaviors in more complex systems – especially ones that are both highly adhesive and cohesive – are poorly understood.

Despite the continued interest in texture-taste interactions for more than 50 years, the specific mechanisms and interactions are still unclear. In order to translate this knowledge into design rules for food manufacturing, several important questions should be addressed. For example, the role of mucoadhesion in taste perception has seen little attention in the literature. Mucoadhesion is a well-known phenomenon in the medical field, where it is leveraged for controlled drug delivery of active ingredients to mucosal or epithelial cells (Smart, 2005). Many of the hydrocolloids used in taste-texture studies can be classified as mucoadhesives, most notably carboxymethyl cellulose. Vis-à-vis taste perception, persistence on oral surfaces could theoretically lengthen the total duration of sweet taste and consequently the perceived sweetness.

Additionally, the role of microstructure in taste perception – beyond simply the critical overlap concentration – is unknown. Taste suppression above c* is well-established, but the effects of microstructure at concentrations below c* are unclear. This is especially important considering that c* corresponds to zero shear viscosities of 22.0–55.2 mPa·s for guar gum, HPMC, and λ–carrageenan (Cook et al., 2003). If taste suppression only occurs at viscosities above this, then many fluid foods would have polymer usage levels below c*. Moreover, the effects of c* in the literature only apply to simple polymer-thickened aqueous systems. The inclusion of proteins and fat may disturb the overlapping polymer network and reduce inefficient mixing in the oral cavity. This could be leveraged in an emulsified or micro phase-separated system to increase tastant concentration in the continuous or serum phases to facilitate mass transport to receptors. Future studies should consider if microstructure can be altered to control mass transfer of tastants during oral processing.

Based on these gaps, I propose the following hypotheses, which will be addressed in subsequent chapters. I hypothesize that oral processing parameters are adjusted to manipulate underlying food structure, with different structures and food compositions exhibiting unique oral processing fingerprints. This will be assessed utilizing a complex model system and by evaluating food structure, sensory texture, and oral processing parameters. This approach will

be expanded to test the hypothesis that adhesiveness is a dynamic property influenced by both degree of viscoelasticity and interfacial energy between food and contact surface.

Specifically in regards to taste perception, I propose that food structure has an effect on temporal attributes of both taste and texture. Moreover, due to multi-modal sensations of texture, different sweeteners (both nutritive and non-nutritive) will exhibit different temporal behaviors. This study will be designed around a model system where texture is modified without adding ingredients or altering macromolecular profile. Second, I propose that the critical overlap mechanism explaining taste suppression in polymer solutions does not apply to complex food systems due to underlying differences in microstructure. This is an important missing piece in trying to relate previous taste-texture studies that used simple model systems to complex food systems. The last research focus is on mucoadhesion, where to date no studies have specifically evaluated the role of mucoahdesion in temporal taste perception.

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CHAPTER 2. DESIGNING FOODS FOR SATIETY: THE ROLES OF FOOD STRUCTURE AND ORAL PROCESSING IN SATIATION AND SATIETY

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Designing foods for satiety: The roles of food structure and oral processing in satiation and satiety

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ABSTRACT

Food consumption is determined by a range of factors that contribute to satiation, which ends a meal, and satiety, which determines time between meals. Food structure and texture contribute to satiation and satiety; however, the precise mechanisms are not fully established. The time required for oral processing has been shown to influence satiation/satiety, but the roles of physiological elements of oral processing, such as muscle activity, jaw movement, and tongue movement, remain to be established. Relationships among food structure, texture, oral processing, and satiation/satiety are discussed in reference to designing foods to maximize the contribution of food structure to satiation/satiety.

Contents

1. Introduction

A growing world population combined with global climatic changes has urged society to critically evaluate the agriculture-food continuum. The relatively new concept of "food security" is a primary consideration in order to feed 9 billion people by 2050 (Godfray et al., 2010). Ideally, this would be accomplished by sustainable increases in agricultural production with foods that supply the ideal amount of nutrients – preventing the problems of insufficient nutrients and also inhibiting the over consumption of nutrients. The latter is an immediate problem manifested in obesity rates of 36% in the US and 13% globally (Weight-Control

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Information Network, 2012; World Health Organization, 2015). While the causes of obesity are complex, one approach to curb obesity is to develop tactics that decrease our daily consumption of food. Our food preferences and consumption habits can be better understood by analyzing the roles played by food structure and individual nutrients, which are driven by our choices in agricultural production.

A general food consumption model consists of three daily meals and possible intermittent snacks. The cessation of a meal occurs when a point of *satiation* is reached, whereas the time between meals is a measure of *satiety*. The amount of food consumed during a day, controlled in part by satiation and satiety, should supply nutrients responsible for health and energy in balance with energy expenditure. This is a simplistic representation of a very complex process; nonetheless, it is clear that foods that increase satiation and satiety should be helpful in controlling food intake. Multiple variables influence the onset of satiation and satiety; therefore, designing foods that provide early satiation and enduring satiety requires the consideration of overlapping interactions among food composition, food structure, oral processing, and dynamic sensory perception as well as psychological inputs such as environment and hedonic liking.

In the context of food consumption from mouth through digestion, one could hypothesize that perceptions of satiation and satiety are formed by: 1) specific types of food molecules, 2) specific food structures and their transformations during oral processing, 3) physiological oral processing activities required for bolus formation, 4) oral processing time, or 5) a combination thereof. Though psychological inputs are also of great importance, the goal of this review is to discuss the role of food structure and oral processing in satiation/satiety and to evaluate the current level of mechanistic understanding. Recent reviews have addressed the relationships among food structure, oral processing, and sensory perception (Foster et al., 2011; Koç, Vinyard, Essick, & Foegeding, 2013; Pascua, Koç, & Foegeding, 2013; Stieger & van de Velde, 2013), sensory perception and satiation/satiety (de Graaf & Kok, 2010; de Graaf, 2012), and chewing, appetite, food intake, and gut hormones (Miquel-Kergoat, Azais-Braesco, Burton-Freeman, & Hetherington, 2015). We build on those reviews and add the potential contribution of physiological processes associated with oral processing.

2. Building food structure

In order to discuss structural breakdown and changes during oral processing (and how they may influence psychological and physiological aspects of perception), it is important to first consider the nature of food structure and texture. From the perspective of states of matter, van Vliet, van Aken, de Jongh, and Hamer (2009) classified foods as fitting into broad categories of: fluids, semisolids, soft solids, and hard solids. While universal delineations among categories are difficult, a first approximation is that: fluids flow and have minimal or no yield stress (e.g., beverages); semisolids are fluid-like, have a high yield stress, and deform or break without fracturing into pieces (e.g., pudding, yogurt, and bananas); soft solids fracture into pieces but without sound (e.g., cooked egg white, some cheeses and processed meats); and hard solids fracture into pieces and emit sound during fracture (e.g., crackers, toast, nuts, apples, and carrots). These differences in overall physical properties are due to elements of food structure. The assembly of molecules into food structures occurs by biological processes (i.e., plant and animal cells and tissues), food processing (e.g., processed cheese, gummy bears, bread, pasta, and tofu), or a combination of biological processes and food processing (e.g., whipped cream, pepperoni, and orange juice). Food structures can also be composed of multiple biological and processed components, such as yogurt containing oats or fruit pieces.

The diversity of food structures provides for culinary delight but greatly complicates our understanding of the contribution of structure to satiation/satiety. Foods based on biological structures, such as apples and carrots, are useful because they are part of a normal diet; however, they exhibit inherent variability among samples. This cannot be adjusted and can only be controlled by rigorous selection. In contrast, the structure of foods formed during processing is more easily controlled. Model foods are designed as simplified food analogs that reflect key elements of normal food. For example, biopolymer gels have been used extensively as models for soft solid foods (Çakir, Daubert et al., 2012; Çakir, Vinyard et al., 2012; Devezeaux de Lavergne, van Delft et al., 2015; Devezeaux de Lavergne, van de Velde et al., 2015; Hayakawa et al., 2014; Ishihara et al., 2011; Koç et al., 2014; Kohyama et al., 2015, 2016). However, compared to the general complexity of everyday foods, model systems may limit the interpretation of experimental results. The relative merits of different structures in investigating the contribution of food structure to satiation/satiety will be discussed in subsequent sections.

3. Food structure and oral processing

Foods can be considered a collection of molecules formed into structures that are transformed into a bolus during oral processing. Food oral processing encompasses a range of variables, including oro-sensory time (de Graaf & Kok, 2010; Haber, Heaton, Murphy, & Burroughs, 1977; Hogenkamp & Schiöth, 2013), muscle and jaw activity (Agrawal, Lucas, Bruce, & Prinz, 1998; Brown, Eves, Ellison, & Braxton, 1998; Çakir, Koç et al., 2012; Ishihara et al., 2011; Koç et al., 2014; Peyron, Lassauzay, & Woda, 2002; Smit, Kemsley, Tapp, & Henry, 2011), tongue behavior (de Wijk, Engelen, & Prinz, 2003; de Wijk, Polet, Bult, & Prinz, 2008), bite size (Bolhuis et al., 2014; de Wijk et al., 2008; Forde, van Kuijk, Thaler, de Graaf, & Martin, 2013; Spiegel, 2000), and palatability (Bellisle, Guy-Grand, & Le Magnen, 2000; de Wijk et al., 2008). Oral processing parameters are directly influenced by, and adjusted to accommodate, changes in food texture throughout the chewing sequence (Hiiemae et al., 1996; Mioche, Bourdiol, Martin, & Noël, 1999; Peyron et al., 2002). Additionally, texture perception, which influences expectations of satiety, is a dynamic process influenced by oral processing (Hiiemae, 2004). Thus, the interrelations among food structure, structural breakdown during oral processing, and sensory perception of texture are all important in understanding the effects on satiation and satiety.

In the simplest sense, oral processing can be considered the specific movements of the tongue and jaw – and the muscle activities required for these movements – during manipulation of food in the mouth. These motions are controlled by a central pattern generator in the brain stem (Yamada, Yamamura, & Inoue, 2005); thus, feedback mechanisms regulate the oral process and adjust to changes in food texture and sensory perceptions. Lund (1991) divided the oral process into three phases: preparatory cycles, particle reduction cycles, and pre-swallowing cycles (Lund, 1991). These divisions are reflected in the transport model of Hiiemae and Palmer, which describes the three dynamic stages associated with mastication (Hiiemae & Palmer, 1999). Stage I occurs immediately after a bite of food, where the bite is moved by a series of low amplitude, simple movements of the jaw without occlusion of the teeth. The duration of this step is dependent on the textural attributes of the food material. During stage II, the bite is transformed into a bolus through comminution of the material combined with lubrication from saliva. Depending on the type of food material, jaw movement during this phase is often rhythmic with discrete opening, closing, and occlusal phases. Textural

perceptions of food particle size, viscosity, saliva incorporation, and food bolus cohesiveness/adhesiveness determine at which point the bolus can be safely swallowed (Brown et al., 1998; Engelen, Fontijn-Tekamp, & van der Bilt, 2005; Lucas, Prinz, Agrawal, & Bruce, 2002; Lucas, Prinz, Agrawal, & Bruce, 2004). During stage III, the bolus is transported to the back of the tongue and swallowed (Heath & Prinz, 1999). Hutchings and Lillford (1988) offered one of the earliest models for relating oral processing to texture perception. Their model describes oral processing based on 1) structural breakdown, 2) lubrication of the food bolus with saliva, and 3) time. The underlying principle behind the Hiiemae and Palmer and Hutchings and Lillford models is the transformation of an *initial food structure* into a *bolus structure* with properties that initiate swallowing.

Physiological, neurobiological, and dental oral processing studies measure jaw, tongue, and muscle activities using techniques such as electromyography (EMG), electrognathography (i.e., jaw-tracking), vibromyography (VMG), ultrasonic echo-sonography, or videofluorography. Combinations of EMG and jaw-tracking are most frequently used to investigate how oral processing changes as foods are consumed and texture is assessed (Woda, Foster, Mishellany, & Peyron, 2006). Transformations in structure and composition from initial food structure to a bolus can be tracked via "chew and spit" studies or by methods such as time intensity and temporal dominance of sensations (TDS), which monitor changes in sensory perception throughout oral processing (Jalabert-Malbos, Mishellany-Dutour, Woda, & Peyron, 2007; Mioche, Bourdiol, & Monier, 2003; Pineau et al., 2009).

3.1. Effects of food structure on eating rate and duration

Investigations into oral processing activities pertinent to food structure/texture can be grouped into two general types: studies that use a variety of common foods purchased from a retailer and studies that design, manipulate, or control the food texture of a set of samples. Studies of the former allow inferences to be made across a structural spectrum at the expense of control over specific textural variables. Alternatively, studies of the latter allow for the control of sample nutrient content, volume, structure, and texture, but may not be directly representative of foods consumed on a daily basis or of the combination of multiple textures within one meal.

Oral processing literature to date comprises a wide range of food products and textures, thus allowing overarching trends to be elucidated. However, to our knowledge no single study has evaluated muscle and/or jaw activities during oral processing of foods spanning the entire range of fluids, semisolids, soft solids, and hard solids. As shown in Table 1, the majority of food oral processing studies based on EMG, VMG, or jaw tracking utilized soft and hard solid structures, such as cheeses, biopolymer gels, or confections. Soft solids are ideal test matrices because 1) a single structural/textural variable is easily manipulated/isolated across a set of samples (e.g., hardness) and 2) soft solids require discrete chews that can be clearly quantified by monitoring muscle and jaw movements. Whether the sample set is purchased or formulated, oral processing studies require a sample set that exhibits a range of intensities of the variable of interest (e.g., soft to hard) in order to determine the effect of one variable upon oral processing behaviors.

Agrawal et al. (1998) were one of the first to demonstrate the dependence of oral processing on intrinsic properties of food texture. Using a selection of cheeses, nuts, and carrots, the authors established a clear dependence of breakdown rate and activity of closing muscles on mechanical parameters – in this case, toughness and modulus of elasticity. More recent studies have expanded on this concept by utilizing a wider variety of foods varying in texture from fluids to soft and hard solids (Forde et al., 2013; Viskaal-van Dongen, Kok, & de Graaf, 2011). Though the foods were not characterized structurally, both Forde et al. (2013) and Viskaal-van Dongen et al. (2011) found significant differences in eating rates, ranging between 4 g/min for crisp foods such as rice cakes or tortilla chips, 100 g/min for soft cooked vegetables, and 650 g/min for thin beverages such as juice or soda. The total number of chews necessary to process 50 g samples ranged from 27 to 488 chews for mashed potatoes and tortilla chips, respectively, while total oral processing time ranged from 27 s for canned tomatoes to 350 s for tortilla chips. Though chewing rates stayed relatively constant across all foods (1 chew/s), faster eating rates were associated with increased food intake (by weight) and high water content foods. This real-world, real-food approach indicates that foods requiring limited oral processing actions, such as liquids, semisolids, and soft, cooked solids, may be more easily over consumed as compared to hard or chewy solids, such as crackers, chips, or certain meats. It is important to mention, however, that these studies measured eating rate by weight (g/min) rather than by total calorie intake (calories/min): food pairs with similar textures and almost identical eating rates also varied greatly in caloric and macronutrient content (e.g., raw carrots and

Table 1
Oral processing studies organized by food texture.[a]

	Fluids	Semi-solids	Soft solids	Hard solids
Biological structure	–	–	beef[1]	apple[2], coconut[3], carrots[4], nuts[5]
Processed structure	–	mayonnaise[6], custard[7]	cheese[8], confections[9], biopolymer gels[10], rice[11], sausage[12]	biscuits[13]
Combination	–	–	–	–

[a] This list only includes studies that utilized electromyography, vibromyography, tongue movement, and/or jaw-tracking to quantify oral processing.
[1] Braxton et al., 1996; Brown et al., 1998; Kohyama & Mioche, 2004; Mathevon, Mioche, Brown, & Culioli, 1995; Mioche et al., 2003.
[2] Brown et al., 1998; Ioannides et al., 2009; Kohyama & Mioche, 2004; Kohyama, Nakayama, Watanabe, & Sasaki, 2005.
[3] Mioche et al., 1999.
[4] Agrawal et al., 1998; Brown et al., 1998.
[5] Agrawal et al., 1998; Kohyama & Mioche, 2004.
[6] de Wijk et al., 2003.
[7] de Wijk et al., 2003, 2008.
[8] Agrawal et al., 1998; Çakir, Koç et al., 2012; Jack, Piggott, & Paterson, 1993; Kohyama & Mioche, 2004; Melito, Daubert, & Foegeding, 2013; Mioche et al., 1999; Yven et al., 2012.
[9] Çakir, Koç et al., 2012; Foster et al., 2006; Lassauzay, Peyron, Albuisson, Dransfield, & Woda, 2000; Mioche et al., 1999; Peyron et al., 2002.
[10] Çakir, Daubert et al., 2012; Çakir, Vinyard et al., 2012; Devezeaux de Lavergne, van Delft et al., 2015; Hayakawa et al., 2014; Ishihara et al., 2011; Koç et al., 2014; Kohyama et al., 2015, 2016.
[11] Kohyama, Yamaguchi et al. (2005); Kohyama & Mioche, 2004.
[12] Devezeaux de Lavergne, Derks, et al., 2015; Mioche et al., 1999.
[13] Brown et al., 1998.

steak pieces; mashed carrots and lasagna) (Forde et al., 2013). Though eating rate is important in understanding the effects of food structure upon food consumption, one must acknowledge that raw vegetables and potato chips, eaten at the same rate, will provide a different number of calories per minute. Thus, we cannot assume that a slower eating rate always leads to lower caloric consumption.

3.2. Effects of food structural/textural attributes upon oral processing

Food hardness is historically the most common and reliable variable used when isolating the effects of specific textural variables upon oral processing. Since sensory hardness is scored as a resistance to deformation or effort to fracture, it can be due to a wide range of food structures. There is generally a strong correlation between instrumental and sensory measures of hardness (Barrangou, Drake, Daubert, & Foegeding, 2006; Brown, Foegeding, Daubert, Drake, & Gumpertz, 2003; Kohyama et al., 2016). Across a wide variety of foods (including biopolymer gels, cheeses, confections, meats, fruits, and vegetables), increasing food hardness is shown to: 1) increase muscle activity during chewing (Agrawal et al., 1998; Çakir, Koç et al., 2012; Foster, Woda, & Peyron, 2006; Ishihara et al., 2011; Koç et al., 2014; Mioche et al., 1999, 2003; Peyron et al., 2002), 2) increase the duration of oral processing (Çakir, Koç et al., 2012; Foster et al., 2006; Ishihara et al., 2011; Mioche et al., 1999), 3) increase the number of chews required to prepare food for swallowing (Çakir, Koç et al., 2012; Foster et al., 2006; Koç et al., 2014; Peyron et al., 2002), and 4) increase the amplitude of jaw movement in the vertical and medial lateral planes (Koç et al., 2014; Peyron et al., 2002). Structural transformations in soft and hard solids associated with sensory hardness are the formation of particles (first bite) and reduction of particle size (number of chews).

In addition to hardness, other food attributes such as deformability, adhesiveness, and moisture release influence oral processing (Çakir, Koç et al., 2012; Çakir, Vinyard et al., 2012; Foster et al., 2006; Koç et al., 2014). Moreover, they may influence jaw movement in different ways. For example, jaw movement increased in all three planes of movement (vertical, medial-lateral, anterior-posterior) during oral processing of biopolymer gels with greater fracture stress (i.e., hardness) (Koç et al., 2014). Gels with increased fracture strain (i.e., deformability), on the other hand, decreased jaw movement in the vertical and lateral directions and had no effect upon anterior-posterior jaw movement. Increased adhesiveness of caramels required more jaw movement in all three planes and more muscle activity even at equal compressive hardness (Çakir, Koç et al., 2012).

3.3. Effects of structure on dynamic oral processing and sensory perception

Oral processing behavior is sensitive to specific structural differences within textural categories of soft and hard food solids. Brown et al. (1998) differentiated cell-based crunchy and juicy hard solids (apples, carrots) from crisp and dry hard solids (shortbread biscuits) based on chewing patterns. Chewing crunchy and juicy foods required vertical compression that decreased in effort over time, while processing of crisp and dry biscuits began with vertical compression and transitioned into shearing movements with the teeth with low tooth contact at each chew. Different microstructures of protein-polysaccharide gels are associated with distinct oral processing patterns (Çakir, Vinyard et al., 2012). Comparisons of low and regular fat cheese and caramels indicate that, regardless of sample hardness, decreases in fat content increase the number of chews, chewing duration, and muscle activity during consumption. Foster et al. (2006) highlighted the differences between confections with elastic or plastic textures. Though both plastic caramels and elastic gelatin gels required the same number of chews, the caramels slowed chewing rate and increased vertical and lateral jaw movements (Foster et al., 2006).

In addition to manipulating viscosity (food structure) and fat percentage (nutrient content and food structure), de Wijk et al. (2008) examined the effects of incorporating different sized particles into vanilla custards. Larger particle sizes increased overall muscle activity during chewing while increasing lipid content decreased digastric muscle activity associated with swallowing (de Wijk et al., 2008). Perception of specific sensory attributes were associated with particular oral behaviors: increasing viscosity and melting sensations were associated with increased overall muscle activity while jaw opening and closing were related to lower ratings of sample thickness. Additionally, ratings of liking were associated with oral movements during the first 1–2 s of oral processing, whereas creaminess ratings related to movements 4–5 s into consumption. The authors concluded that foods have individual oral processing "fingerprints" that determine time-dependent sensory perceptions.

As observed by de Wijk et al. (2008), initial food structure, individual chewing behavior, and dynamic changes in food texture throughout oral processing impact the trajectory of sensory perception. Sensory perception is altered by input of chewing energy, chewing pattern, chewing rate, and tongue movement (Braxton, Dauchel, & Brown, 1996; Brown & Braxton, 2000; Devezeaux de Lavergne, Derks, et al., 2015; de Wijk et al., 2003). In particular, work from both de Wijk et al. (2003) and Devezeaux de Lavergne, Derks, et al. (2015) indicates that individual hedonic preferences for specific sensations or sensation intensities may shape individual oral processing patterns.

4. Satiation and satiety

Determining the role of food structure in satiation and satiety is difficult because these sensations are impacted by physical, physiological, and psychological factors before, during, and after eating. These include the specific eating environment (Wansink, 2010), food appearance or labeling (Crum, Corbin, Brownell, & Salovey, 2011), aroma, volume, and texture (de Wijk et al., 2008; Hogenkamp, Stafleu, Mars, Brunstrom, & de Graaf, 2011; Turgeon & Rioux, 2011; Zijlstra, Wijk, Mars, Stafleu, & Graaf, 2009), bite size (de Wijk et al., 2003; Hogenkamp, Mars, Stafleu, & Graaf, 2010; Hutchings et al., 2009; Weijzen, Smeets, & de Graaf, 2009; Weijzen, Liem, Zandstra, & de Graaf, 2008) consumption time (de Wijk et al., 2008; Zijlstra et al., 2009), digestive speed (Kong & Singh, 2008), and physiological biomarkers (de Graaf, Blom, Smeets, Stafleu, & Hendriks, 2004). Changes in nutrient profile and food structure also significantly impact oro-sensory time, or the time food spends in the mouth during mastication. Oro-sensory time is the same as oral processing time, with oro-sensory time often used to differ between sensory signals coming from outside the mouth (e.g., sight) with those observed during oral processing. Foundational reviews by de Graaf (2012) and Hogenkamp and Schiöth (2013) thoroughly covered the effects of variables including food texture, bite size, eating rate, and method of consumption upon satiation and satiety, and thus, will not be recapitulated in the current review. One challenge in determining the role of specific oral processing operations is the confounding nature of oro-sensory time. For example, increased hardness should require additional muscle activity and can change jaw movements, but will also require additional oro-sensory time. Very few satiation and satiety studies control or adjust for oro-sensory time, so conclusions therein must be accompanied by this caveat.

4.1. Indicators of satiation and satiety

Measurements of satiation and satiety typically fall into three categories: subjective panelist ratings, physiological measurements, and *ad libitum* intake. Subjective panelist ratings of hunger and fullness, often in the form of a 100 mm visual analog scale (VAS), represent the most direct and simplistic measurement of hunger and fullness. These ratings are often corroborated by offering an *ad libitum* snack or meal and measuring how many calories the panelist consumes following consumption of a test food. One can also measure physiological biomarkers associated with metabolism, satiation, and satiety, including glucose, insulin, ghrelin, leptin, cholecystokinin (CCK), glucagon-like peptide-1 (GLP-1), peptide YY (PYY), and gastric inhibitory peptide (de Graaf et al., 2004). Other physiological measurements include diet induced thermogenesis (Crovetti, Porrini, Santangelo, & Tesolin, 1997; Westerterp-Plantenga, Rolland, Wilson & Westerterp, 1999) and gastric emptying time (Blundell et al., 2010). While quantification of biomarkers provides an unbiased, physiological response, healthy panelists often exhibit wide ranges of biomarker concentrations and this biological variation can complicate interpretation of results. Combinations of the above measurements are more commonly utilized to account for biological and psychological variability.

In addition to *how* satiety is measured, the timing and methodology by which the test food is consumed also complicates results (Livingstone et al., 2000; de Castro, 2004). Test foods can be consumed as a calorie and volume-controlled preload (i.e., snack or appetizer), an *ad libitum* preload, as a meal, or accompanying a meal. Optimally, test food consumption should reflect real world eating situations; however, it must also be realized that testing moves the eating experience from the subconscious to the conscious, adding yet another variable.

4.2. Effects of macromolecules on satiation/satiety

Food is first and foremost classified by its macromolecular and nutritional content, and the specific effects of constituent molecules found in foods are a fundamental starting point in understanding satiation and satiety. Studies comparing the satiating effects of macromolecules are conclusive: protein is the most effective in prolonging feelings of fullness, followed by carbohydrates and then lipids (Bellissimo & Akhavan, 2015; Halton & Hu, 2004; Rolls, Hetherington, & Burley, 1988; Westerterp, 2004). Increasing the daily percentage of dietary protein consumption has been shown to 1) increase diet induced thermogenesis (Westerterp-Plantenga, Rolland, Wilson, & Westerterp, 1999), 2) decrease *ad libitum* food consumption (Brennan et al., 2012; Eisenstein, Roberts, Dallal, & Saltzman, 2002; Latner & Schwartz, 1999), 3) increase satiety ratings (Brennan et al., 2012; Eisenstein et al., 2002; Latner & Schwartz, 1999; Stubbs, van Wyk, Johnstone, & Harbron, 1996; Westerterp-Plantenga et al., 1999), and 4) increase weight loss and/or lower body fat percentage (Lejeune, Westerterp, Adam, Luscombe-Marsh, & Westerterp-Plantenga, 2006; Pasiakos et al., 2013; Veldhorst et al., 2008; Veldhorst, Nieuwenhuizen, Hochstenbach-Waelen, van Vught et al., 2009; Weigle et al., 2005). Induction of satiety by protein is driven by a number of physiological factors resulting from protein digestion, including increased energy expenditure (Westerterp-Plantenga et al., 2006), release of satiety biomarkers such as GLP-1 and CCK (Hall, Millward, Long, & Morgan, 2003; Lejeune et al., 2006; Veldhorst, Nieuwenhuizen, Hochstenbach-Waelen, Westerterp et al., 2009; Westerterp-Plantenga et al., 1999), and regulation of insulin signaling and muscle synthesis (Mellinkoff, Frankland, Boyle, & Greipel, 1997; Morrison, Xi, White, Ye, & Martin, 2007).

Inherent differences in amino acid composition, molecular structure, and enzyme activity significantly affect digestion and absorption speed of proteins (Inglingstad et al., 2010). "Fast" digesting proteins such as whey, soy, and pea increase plasma amino acid concentrations and satiety hormone release earlier than "slow" digesting proteins such as casein or egg albumin that precipitate under the acidic conditions of the stomach and delay gastric emptying (Abou-Samra, Keersmaekers, Brienza, Mukherjee, & Macé, 2011; Anderson, Tecimer, Shah, & Zafar, 2004; Diepvens, Häberer, & Westerterp-Plantenga, 2007; Hall et al., 2003; Pal & Ellis, 2010; Veldhorst, Nieuwenhuizen, Hochstenbach-Waelen, van Vught et al., 2009). However, the selection of specific animal or plant protein sources does not appear to be as important to digestion rate as the total amount of protein consumed (Aldrich et al., 2011; Bellissimo & Akhavan, 2015; Lang et al., 1998). It is important to note that protein digestion is a complex area beyond the scope of this review; readers are directed to the reviews of Mackie and Macierzanka (2010), Lundin, Golding, and Wooster (2008), and Bendtsen, Lorenzen, Bendsen, Rasussen, and Astrup (2013) for additional information regarding protein digestion.

Beyond protein, non-digestible carbohydrates such as fiber and resistant starches found in whole grains, legumes, fruits, and vegetables also impact satiation and satiety when administered both as whole foods and in supplemental form (Lundin et al., 2008; Slavin & Green, 2007). Fiber is shown to induce satiation and satiety by increasing chewing time; increasing secretion of saliva, gastric juice, and satiety hormones; slowing nutrient absorption rate in the small intestine; and by modulating plasma glucose (Bellissimo & Akhavan, 2015; Heaton, 1973; Kong & Singh, 2008). However, effectiveness varies widely among fiber type (soluble vs. insoluble), quantity, and delivery method (Klosterbuer, Thomas, & Slavin, 2012; Willis, Eldridge, Beiseigel, Thomas, & Slavin, 2009). Generally, 10 g doses, or 30 g throughout the course of a day, are shown to promote satiety (Benelam, 2009; Slavin & Green, 2007). Fibers with high intrinsic viscosity – such as pectin, β-glucan, psyllium, guar gum, and alginate – are most highly correlated with increases in satiety and reductions in energy intake, possibly stemming from their ability to absorb water and form gels within the gastrointestinal tract (Bellissimo & Akhavan, 2015; Hoad et al., 2004; Paxman, Richardson, Dettmar, & Corfe, 2008; Pelkman, Navia, Miller, & Pohle, 2007; Slavin & Green, 2007). In beverage or semi-solid foods, these viscous fibers may also impact oral processing time and effort.

Micronutrients do not significantly contribute to food structure and oral processing, but recent studies indicate that polyphenolic intake may slow digestion and nutrient absorption. Polyphenolic compounds in foods have been shown to form high affinity, non-covalent bonds with both food proteins and digestive enzymes; this binding is suspected to decrease enzymatic efficiency by changing the three-dimensional protein structure or by interrupting protein receptors or enzymatic binding sites (Jakobek, 2015; Nagy et al., 2012). *In vitro,* tannins in cocoa, fruits, tea, cinnamon, and peanuts have been shown to slow the breakdown of carbohydrates and lipids by inhibiting digestive enzymes (Adisakwattana, Lerdsuwankij, Poputtachai, Minipun, & Suparpprom, 2011; Barrett et al., 2013; Grove, Sae-Tan, Kennett, & Lambert, 2012; Gu, Hurst, Stuart, & Lambert, 2011; McDougall et al., 2005; Nakai et al., 2005; Oboh et al., 2014; Yu, Sun, & Gao, 2014), and higher degrees of polyphenol polymerization are associated with greater potential for inhibition of digestive enzymes (Barrett et al., 2013; Jakobek, 2015). These observations have translated to increased satiety ratings, decreased digestive efficiency, and increases in fecal nutrient excretion in a limited number of *in vivo* studies and clinical trials (Josic, Olsson, Wickeberg, Lindstedt, & Hlebowicz, 2010; Unno et al., 2009; Wang, Noh, & Koo, 2006).

Macro- and micronutrients are the foundation of food structure and metabolism. However, satiation, satiety, and total caloric consumption are impacted by more than the molecules found in foods. Manipulation of the food nutrient profile, such as changing protein or fiber type, introduces inherent variations in food structure, texture, flavor, and appearance. These differences can impact oral processing patterns, oral processing time, and hedonic ratings. As suggested by Halton and Hu (2004), incongruences among studies investigating the effects of food macronutrients on satiation and satiety may be attributed to inconsistencies in food form. Though often overlooked, test foods provided as a nutritional supplement (beverage or pill), mixed meal (whole foods), meal replacer (shake or bar), or as a preload snack significantly change oral processing and affect physiological and psychological satiety.

4.3. Approaches to food texture modification for satiety studies

Investigations into the effects of food texture on satiation/satiety involve either testing a variety of foods or directly modifying texture in specific ways. Texture can be modified by adding an ingredient to increase viscosity or thicken, by breaking down structure through mechanical action, or by incorporating air. The manipulations can either be within a textural category (e.g., thin and thick fluids) or across categories (e.g., fluid to semisolid gel).

A collection of studies evaluating the effects of food texture on satiation and satiety is shown in Table 2. Studies accompanied by a plus symbol (+) indicate that at least one measured marker of satiation/satiety was affected by texture; however, this does not mean that all markers within that category were affected. To date, fluids are the most commonly evaluated texture type and subjective panelist ratings are the most frequently used measurement to quantify satiation and satiety. This is followed by roughly half of the studies measuring *ad libitum* intake (either of the product or of a subsequent meal) and less than a third that used blood or hormone biomarkers. Only one study used all three types of measurements.

4.3.1. Adding an ingredient to thicken fluids and semisolids

The simplest way to modify food texture in fluids and semisolids is by adding an ingredient to increase viscosity. For example, drinkable yogurts, juices, and puddings were rated as more likely to be filling when thickened with tara gum, guar gum, starch, or β-glucan (Bertenshaw, Lluch, & Yeomans, 2013; Mars et al., 2009; McCrickerd et al., 2012; McCrickerd, Chambers, & Yeomans, 2014; Pentikäinen et al., 2014; Zhu et al., 2013). Similar results are seen with vanilla milkshakes of two different viscosity levels, where thickness lowered hunger by a greater magnitude and for a longer period of time (Mattes & Rothacker, 2001).

As expected from the above discussion, the type of ingredient used to thicken likely plays a contributing role in modulating hunger. At two different levels of viscosity, whey protein beverages elicited lower subjective hunger ratings than beverages thickened with sodium alginate, suggesting both molecular and physical impacts on perception of satiety (Solah et al., 2010). A similar approach was taken for isocaloric beverages formulated from thick fluids to semisolids (Hogenkamp, Mars, Stafleu, & de Graaf, 2012; Zijlstra, Mars, de Wijk, Westerterp-Plantenga, & de Graaf, 2008;

Table 2
Studies evaluating the effects of food texture on satiation and satiety.[a]

Texture type(s)	Food(s)	Texture modification	Satiation/Satiety			
			Subjective ratings	*Ad libitum* intake	Biomarkers	Study
fluid	yogurt drink	thicken	+	+		Mars et al. (2009)
fluid	yogurt drink	thicken	+			McCrickerd et al. (2012)
fluid	oat beverage	thicken	–		+	Juvonen et al. (2009)
fluid	beverage	thicken	+		+	Marciani et al. (2001)
fluid	orange juice	thicken	+			Pentikäinen et al. (2014)
fluid	milkshake	thicken	+			Mattes and Rothacker (2001)
fluid	yogurt drink	add air	+	+		Rolls et al. (2000)
fluid	beverage	thicken	+			Solah et al. (2010)
fluid	yogurt drink	thicken	+	+		Bertenshaw et al. (2013)
fluid, semi-solid	dairy beverage, semi-solid	thicken	–	+		Zijlstra et al. (2008)
fluid, semi-solid	dairy beverage, semi-solid	thicken	+		–	Zijlstra et al. (2009)
fluid, semi-solid	dairy beverage, semi-solid	thicken	+	+		de Wijk et al. (2008)
fluid, semi-solid	drinkable yogurt, yogurt	grind	+	–		Tsuchiya et al. (2006)
fluid, semi-solid	drinkable yogurt	comparison	+	+		Hogenkamp et al. (2012)
fluid, semi-solid, solid	apple, applesauce, apple juice	grind	+	+		Flood-Obbagy and Rolls, (2009)
fluid, semi-solid, solid	apple, applesauce, apple juice	grind	+			Mattes and Campbell (2009)
fluid, solid	vegetable soup	grind		–		Flood and Rolls (2007)
fluid, solid	watermelon	comparison	+			Mourao et al. (2007)
fluid, solid	dairy	comparison	+			Mourao et al. (2007)
fluid, solid	coconut	comparison	+			Mourao et al. (2007)
fluid, solid	chicken	grind	+		–	Martens et al. (2011)
fluid, solid	vegetable pie	grind	+		+	Santangelo et al. (1998)
fluid, solid	vegetable pie	grind			+	Peracchi et al. (2000)
fluid, solid	meal replacement	comparison	+	+		Stull et al. (2008)
fluid, solid	meal replacement	comparison	+		+	Leidy et al. (2010)
fluid, solid	meal replacement	comparison			+	Tieken et al. (2007)
semi-solid	pudding	thicken	+	–	+	Zhu et al. (2013)
soft solid	lunch meat	comparison	–	–		Zijlstra et al. (2010)
soft solid	meat substitute	comparison	–	–		Zijlstra et al. (2010)
soft solid	gelatin confections	comparison	–	–		Zijlstra et al. (2010)
soft solid	savory meal	mashed		+		Forde et al. (2013)
soft solid	bread roll	add air	+	+		Irvine et al. (2005)
soft solid	savory meat pie	grind	–	+		Pritchard et al. (2014)
hard solid	breakfast cereal	add air	+	+		Irvine et al. (2007)
hard solid	cheese puffs	add air	–	+		Osterholt et al. (2007)

[a] Studies accompanied by a plus symbol (+) indicate that at least one measured marker of satiation/satiety was affected by texture.

Zijlstra et al., 2009). *Ad libitum* intake of the liquid sample was 30% greater than the semisolid in a real life setting and 29% higher in a lab setting (Zijlstra et al., 2008). Hogenkamp et al. (2010) observed a similar effect in liquid and semisolid yogurts consumed with either a straw or a spoon: no significant differences were observed between foods eaten with a spoon, but the liquid consumed via straw was consumed at a faster rate and elicited higher *ad libitum* consumption. Both studies indicate that viscosity has an effect on *ad libitum* intake, which the authors attributed – at least in part – to faster consumption (i.e., shorter oro-sensory time) of fluids.

Contradictions between psychological and physiological measurements are a complicating factor in satiety testing. For example, higher viscosity beverages appear to delay gastric emptying (Juvonen et al., 2009; Marciani et al., 2001), suggesting a physical mechanism of satiety. However, for low viscosity, fast digesting beverages, Juvonen et al. (2009) observed higher levels of satiation and satiety biomarkers (glucose, insulin, CCK, GLP-1, PYY), while Marciani et al. (2001) only reported changes to subjective hunger. This ambiguity may be explained by a physiological mechanism: the faster digestion and gastric emptying proceed, the faster glucose and amino acids are released into the bloodstream, triggering release of peptides and hormones that elicit satiation. Until more research is available, it appears that a range of food viscosities may be effective in inducing satiety via multiple mechanisms.

4.3.2. Grinding or homogenizing food

Another method used to produce a range of food textures is to grind, purée, or homogenize a solid food structure. In this approach, nutritional content remains constant and panelists consume a specific volume of water to compensate for any an-isovolumetric samples. Fullness ratings after consumption of cell-based natural foods are found to be inversely related to degree of structure. Several studies reported that whole apples were more filling and attenuated hunger more than puréed or juiced apples (Flood-Obbagy & Rolls, 2009; Mattes & Campbell, 2009). An evaluation of solid and liquid versions of high carbohydrate (watermelon), high protein (cheese), and high fat (coconut) foods indicated that total daily energy intake is higher following consumption of the liquid versions of each of these foods, regardless of macronutrient tested (Mourao, Bressan, Campbell, & Mattes, 2007). Similar results were reported for chunky and puréed fruit yogurt compared to fruit dairy beverages and fruit juice and for whole and puréed versions of chicken, vegetable pies, and meat pies (Martens, Lemmens, Born, & Westerterp-Plantenga, 2011; Peracchi et al., 2000; Pritchard, Davidson, Jones, & Bannerman, 2014; Santangelo, Peracchi, Conte, Fraquelli, & Porrini, 1998; Tsuchiya, Almiron-Roig, Lluch, Guyonnet, & Drewnowski, 2006). These findings suggest that oral processing efforts, oral processing time, or a combination of both, may contribute to satiety. However, research supporting the satiety-inducing effects of solid foods is not as conclusive as that for increasing viscosity in fluid and semisolids. Flood and Rolls (2007) and Mattes (2005) reported no differences in hunger ratings or *ad libitum* intake following consumption of chunky or puréed vegetable soups. These discrepancies may be due to the sample calorie content, timing of food consumption, or the effect of uncontrolled oro-sensory time.

Another approach in evaluating texture effects on satiety is the comparison of solid and liquid forms of meal replacement products, such as bars and beverages. In this sense, the beverage is not a puréed version of the solid *per se*, but products coming from the same manufacturer typically target similar nutrient content and macromolecular ratio, thus allowing an *ad hoc* comparison of texture. The solid "bar" versions of meal replacements promote lower postprandial hunger ratings and higher fullness ratings than beverages (Leidy, Armstrong, Tang, Mattes, & Campbell, 2010; Stull, Apolzan, Thalacker-Mercer, Iglay, & Campbell, 2008; Tieken et al., 2007). Moreover, solid versions reduced *ad libitum* intake two hours after the meal (Stull et al., 2008). The effects on physiological markers were less evident, although in both studies by Tieken et al. (2007) and Leidy et al. (2010), a decrease in ghrelin was observed after consumption of the solid variants. Here again, oral processing efforts, oral processing time, or a combination of both, may be contributing to satiety.

4.3.3. Increasing food volume

Beyond differences in texture, satiation and satiety are tied to the volume of a food consumed (Bell & Rolls, 2001). As such, reducing the energy density of food is another potential way to promote satiety. Using water to add volume appears to promote satiation but may not sustain feelings of satiety (Almiron-Roig, Flores, & Drewnowski, 2004; Rolls et al., 1998). Alternatively, the incorporation of air is an effective method of reducing total energy consumption of yogurt beverages, bread, breakfast cereal, and hard-solid snacks (Irvine et al., 2005; Irvine, Livingston, & Welch, 2007; Osterholt, Roe, & Rolls, 2007; Rolls, Bell, & Waugh, 2000). The results of these studies indicate that although a greater volume of food is consumed, total mass and energy consumption is reduced due to low energy density of the high volume food (Osterholt et al., 2007). This is concomitant with a significant increase in reported satiety and decrease in *ad libitum* intake for the aerated (i.e., low energy density) foods (Irvine et al., 2005, 2007). It has also been suggested that sensory-specific satiety – the concept that continued consumption of a product leads to a decrease in acceptance that eventually contributes to satiation – is driven by food volume more than energy content (Bell, Roe, & Rolls, 2003).

4.4. Effects of food structure upon satiety and satiation

The current state of satiety literature suggests that food texture may be an important contributing factor in the control of satiation, satiety, and total daily caloric intake. It is unclear however, whether this effect is direct or indirect, as few studies testing food texture have controlled for oral processing parameters such as oro-sensory time, muscle recruitment, jaw activity, number of chews, or dynamic textural perception. Most studies have focused on comparisons between fluid and semisolid textures or between whole and puréed versions of the same food(s); consequently, there is compelling research that higher viscosity fluids and semisolids may increase ratings of satiety and decrease *ad libitum* intake. The positive correlations between viscosity and satiety may be due to a natural decrease in eating rate, increased tongue and muscle movements, or the requirement of a spoon or straw, all of which extend the time necessary for the brain to process physiological and psychological signals. In addition to structure altering oral processing, structural differences at the microscale may remain in the bolus and modulate satiety during the digestive processes. When consuming emulsion preloads with similar liking and texture but different droplet sizes, the emulsion with smaller droplets resulted in less overall intake of a subsequent lunch (Lett, Norton & Yeomans, 2016). This suggests that structural elements not detected as differences in sensory texture can contribute to satiety.

Significant differences in satiety conferred by soft and hard solids are still not clear and require additional inquiry. Oral processing studies have firmly established that increasing food hardness increases oral processing time, muscle activity, and jaw movement, but these observations have not been linked with satiety in human clinical trials. This is partially due to the difficulties involved in controlling multiple variables when

designing or selecting food products. Additionally, the results of experiments measuring satiety are often dependent on whether the food is provided as a snack, a meal, or in the context of a larger meal. The time of day and timing of test food consumption, satiety ratings, and *ad libitum* meal also play an important role (Livingstone et al., 2000). In a real-world scenario, these variables interact to form psychological expectations and physiological effects, further complicating measured outcomes.

5. Designing foods for satiation and satiety

As shown in Fig. 1, the selection or design of foods to maximize satiation and satiety encompasses multiple considerations. First, composition – with special consideration regarding macromolecules – is selected based on known molecular associations with satiation/satiety and/or the ability to form specific food structures. That food structure is then consumed, during which it is manipulated via oral processing to generate sensory perceptions of texture and flavor. Oral processing behaviors and simultaneous sensory perceptions overlap during mastication, as dynamic sensory perceptions provide feedback that drive changes in muscle activity and associated jaw and tongue movements. Finally, during and after oral processing, the body registers perceptions of satiation and satiety, which are driven by physical, physiological, and psychological factors. Therefore, food structure can be linked with factors influencing satiation and satiety starting with the initial visual assessment of food, through transformations during mastication, and finally with how structural elements affect digestion and absorption.

Literature to date suggests that the most important elements of satiety-inducing foods are: 1) the inclusion of protein (macromolecular, physiological), 2) an increase in food volume (physical), and 3) a structure requiring a long oro-sensory time (physiological, physical, psychological). Numerous studies have provided convincing evidence that protein, regardless of source, has a higher satiating capacity than carbohydrates or lipids (Bellissimo & Akhavan, 2015; Halton & Hu, 2004). Increasing food volume, via the incorporation of water or air, may increase oral processing time, decrease energy density, and/or occupy a greater volume in the stomach and intestines, all of which have been found to increase ratings of satiation and satiety (Bell & Rolls, 2001; Rolls et al., 1998). Finally, the design of a hard or chewy food structure extends oral processing effort, increasing the time for physiological and psychological signals to reach the brain.

It must be specified, however, that longer oral processing time does not always correlate with increased oral processing effort. If a hard candy is allowed to dissolve undisturbed in the mouth, the oral processing time required is long, due to slow dissolution, but very little muscle, jaw, or tongue activity is required. Alternatively, chewy foods must be physically broken down into small particles and formed into a bolus that can be safely swallowed. Oral processing effort is *indirectly* quantified by total amounts of muscle activity and jaw and tongue movement; however, true work (i.e., force per distance) cannot be *directly* measured by EMG or jaw-tracking. Chewiness or the number of chews required before swallowing depends on multiple characteristics including adhesiveness (e.g., caramel), hardness and brittleness (e.g., peanuts), or toughness (e.g., steak, lettuce) that require both long oral processing times and extensive muscle, jaw, and tongue activity. Ideally, foods that encompass all three of these characteristics, such as a chewy protein-based gel, would be expected to have the greatest effect upon satiation and satiety.

Based on oral processing studies to date, designing food structures that increase the hardness, thickness, or toughness of foods in order to increase oro-sensory time would hypothetically increase satiety. However, the crux of this hypothesis lies in the paradox of formulating healthier foods: the removal of fat, sugar, and salt from processed foods often increase hardness or chewiness (e.g., reduced or low fat cheese, rice cakes), but also impacts flavor perception and decreases overall product liking. The oro-sensory time-satiety hypothesis may hold true if flavor and hedonic properties are maintained between foods requiring short

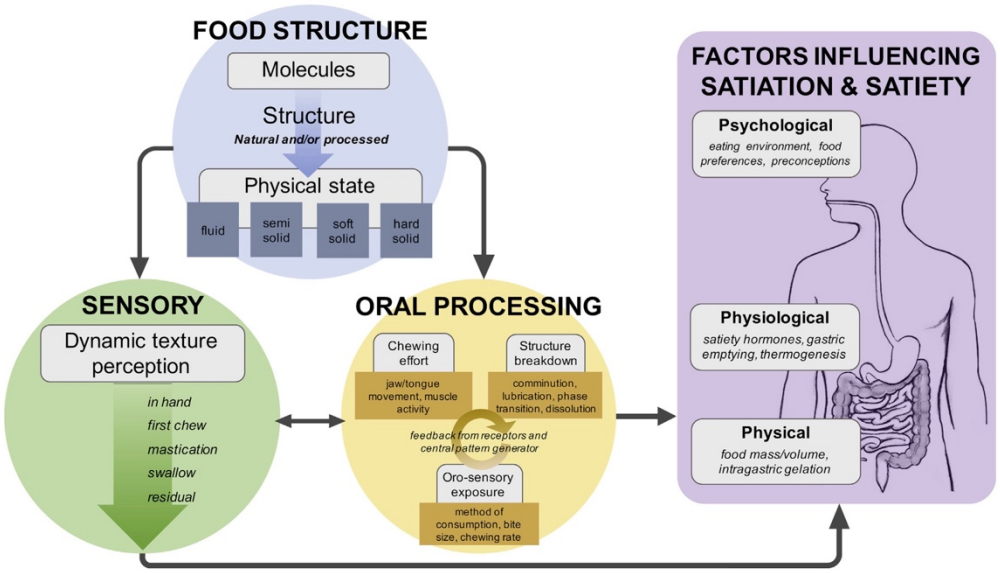

Fig. 1. The relationships among food structure, oral processing, sensory perception, and satiation and satiety.

and long oro-sensory times, but not if product liking is impacted. In fact, consumers may even modify oral processing behaviors to reflect dislike of a healthier or less pleasing product, thereby counteracting the potential benefits of longer oro-sensory time. Additionally, oro-sensory time is likely not the only variable that determines feelings of satiation and satiety. For foods that require the same extent of chewing, such as carrots, caramels, or protein bars, consumption outcomes can also be driven by macromolecular and caloric contents, hedonic liking, and jaw, muscle, or sensory fatigue.

Oral processing behaviors, satiation, satiety, and food consumption are a sum of past experiences, expectations, environment, and hedonic preferences for specific food textures and flavors. Multiple studies have suggested that individuals adjust their oral processing behaviors to optimize or enhance textural or flavor perceptions while eating (Devezeaux de Lavergne, Derks, et al., 2015; de Wijk et al., 2003; Jeltema, Beckley, & Vahalik, 2015). Devezeaux de Lavergne, Derks, et al. (2015) observed that eating speed (fast vs. slow) leads to differences in dynamic textural perception, while Jeltema et al. (2015) hypothesized that individual oral processing behaviors drive preferences for specific food textures. These tailored behaviors may enhance enjoyment during eating, which may carry over to increased ratings of satiety. Alternatively, it is possible that enhanced enjoyment of certain foods via oral processing may also encourage overconsumption of calories during eating, as enjoyment could override satiation signals.

6. Future outlook

To date, food formulation and health research have been clearly divided between those studying the effects of food texture upon either oral processing or upon satiation and satiety. Tenuous conclusions can be drawn between those studies utilizing similar model foods; however, disconnects between these research goals leave major gaps in knowledge. Future satiety studies need to further characterize the structural, rheological, sensorial, and oral processing parameters of foods in order to account for variables beyond nutritional content, volume, and approximated food form. This may require collaboration among research groups to appropriately cover a range of disciplines, but will result in a more thorough understanding of the many variables involved in food consumption.

Few oral processing or satiety studies have standardized sample oral processing times and no studies have designed oral processing procedures that fix oral processing time while varying oral processing actions. Oral processing studies also need to consider factors intrinsic to the material being tested beyond hardness (e.g., adhesiveness, moisture/flavor/aroma release, and means of consumption) and extrinsic factors associated with the human subject, such as gender, age, food preferences, expectations, or the surrounding environment (Kohyama & Mioche, 2004; Wansink, 2010; Woda et al., 2006). Above all, these studies need to incorporate methods that allow researchers to follow dynamic changes in food structure, texture and flavor perception during oral processing to determine how the overall eating process influences satiation/satiety.

Additionally, oral processing and satiety studies have traditionally utilized very different food systems due to methodological constraints: oral processing studies have focused almost completely on soft and hard solids while satiation and satiety studies have investigated foods across the fluid, semisolid, and solid textural spectrum. Because fluids and semisolids require little or irregular jaw movement, oral processing studies will need to incorporate methods to track tongue movement in addition to jaw movement and muscle activity. Feasibility issues and difficulties in data analysis have minimized the measurement of tongue movement in oral processing studies. However, a limited number of tongue movement methodologies exist, most found in the field of speech therapy and articulation. To quantify tongue movements, research groups have utilized multi-point force sensors attached to the upper palate (Hayashi et al., 2013; Sasaki et al., 2013; Yokoyama et al., 2014), EMG electrodes attached to lingual muscles on the underside of the jaw (Sasaki et al., 2013), or by attaching a magnet to the tongue and tracking changes in its magnetic field (Dromey, Nissen, Nohr, & Fletcher, 2006). Development of non-invasive tongue measurement techniques will help to elucidate many of the questions surrounding oral processing of liquids and semi-solids.

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CHAPTER 3. SWEETNESS PERCEPTION IN PROTEIN-POLYSACCHARIDE BEVERAGES IS NOT ALTERED BY VISCOSITY OR CRITICAL OVERLAP CONCENTRATION

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1. ABSTRACT

It is generally reported that in random coil polysaccharide solutions, increasing polymer concentration above the critical overlap concentration (c*) results in decreased taste perception due to a high degree of polymer entanglement and resultant poor mixing efficiency with saliva in the mouth. However, these reports are commonly based on aqueous solutions of polymers and it is unknown if this phenomenon applies to more complex fluids containing protein and fat, or if taste suppression can be explained on the basis of an altered temporal profile of taste. In this study, the effects of carboxymethyl cellulose (CMC) concentration (0.15–1.50% w/w) on sensory texture and sweet taste perception (6.0% w/w sucrose) were evaluated. Zero-shear viscosity of CMC solutions ranged from 5.5 to 133 mPa·s, and c* was experimentally determined to be 0.67% w/w. Three concentrations above c* were tested and only the highest (1.5% CMC) caused a significant ($p < 0.05$) reduction in sweet taste. Moreover, when combined with milk protein concentrate to approximate the macromolecular profile of reduced fat milk, perceived sweet taste increased with viscosity ($p < 0.05$). Time intensity evaluations revealed a large variation in temporal perception of sweet taste among individual panelists, with time to maximum intensity ranging from 4.8 to 33.5 s. Despite a high degree of sensory mucoadhesion, increasing CMC concentration did not significantly alter most time intensity attributes. The results indicate that the critical overlap taste suppression observed in aqueous solutions did not apply to a more complex fluid microstructure containing a combination of protein and fat.

2. INTRODUCTION

Food structure is an important part of the eating process. Viscosity and texture have health implications, where more viscous beverages are reported to attenuate subjective ratings of hunger and reduce *ad libitum* intake (de Wijk, Polet, Bult, & Prinz, 2008; Mars, Hogenkamp, Gosses, Stafleu, & De Graaf, 2009). Hydrocolloids are commonly added to beverages to adjust viscosity, modify texture, or to enhance stability. Despite these functional benefits, the addition of these ingredients can have deleterious effects on taste and flavor perception. Understanding how these ingredients modify taste perception has long been of interest, beginning with seminal studies in the 1970s (Arabie & Moskowitz, 1971; Pangborn,

Trabue, & Szczesniak, 1973). However, the underlying mechanisms and practical implications of taste suppression are still not well understood.

Mathematical and biomechanical models of tastant transfer in the mouth have been developed to understand how food structure influences taste perception (Aubert, Lima, & Le Révérend, 2016; de Loubens, Magnin, Doyennette, Tréléa, & Souchon, 2011; Le Révérend, Norton, & Bakalis, 2013; Pfeiffer, Boulton, & Noble, 2000). From a mass transfer perspective, the oral cavity has been modeled as two regions: an infinitely mixed region comprising the bulk oral cavity and a non-mixing, static salivary layer (i.e., pellicle) on the oral mucosa. A concentration gradient exists between the two regions where mass transfer is governed by Fickian diffusion (Aubert et al., 2016; Kokini, 1987; Le Révérend et al., 2013; Pfeiffer et al., 2000). Recent models have also proposed that due to the rough surface of the papillae, the stagnant salivary layer near receptors experiences low shear rates, which may explain the reported correlations of low shear viscosity and taste perception (Aubert et al., 2016; He, Hort, & Wolf, 2016; Koliandris et al., 2010).

In general agreement with most taste perception models, ingredients or structures that alter the rate and concentration of tastant molecules reaching receptors should alter perceived taste. One early hypothesis explained taste suppression on the basis of decreased diffusion rates with increasing viscosity (Kokini, Bistany, Poole, & Stier, 1982). Although recent models and experimental data both indicate a probable role of restricted diffusion in taste perception, it is likely not the only contributor to such marked reductions in taste that have been reported (Cook, Hollowood, Linforth, & Taylor, 2002; Han et al., 2014; Le Révérend et al., 2013).

Alternatively, the underlying composition and microstructure of a food may play a role in how tastants are perceived during oral processing that involve mechanisms other than restricted diffusion. Baines & Morris (Baines & Morris, 1987) reported taste suppression in random coil polymer solutions that corresponded to the critical overlap concentration, c*. At polymer concentrations above c*, molecular interactions and overlapping hydrodynamic volumes lead to a high degree of polymer entanglement and thus increased viscosity of the system. The authors hypothesized that in this overlapping system, inefficient mixing of solution with saliva results in perceptual changes indicating that this effect is not due to viscosity alone (Baines & Morris, 1987). Taste suppression above c* has been reported for a

number of polymers, including carboxymethyl cellulose, hydroxypropyl methyl cellulose, guar gum, and carrageenan (Baines & Morris, 1987; Cook et al., 2002; Cook, Hollowood, Linforth, & Taylor, 2003; Han et al., 2014; Hollowood, Linforth, & Taylor, 2002). Additionally, the concept of inefficient mixing of saliva and sample above c* implies that pockets of tastants are physically obstructed and delayed from reaching receptors. This may indicate that taste intensity reduction is due to altering the temporal pattern of tastant release associated with changes in food microstructure; however, no studies have specifically evaluated temporal taste patterns at hydrocolloid concentrations above and below c*.

The bulk of the literature agrees with taste suppression when the polymer concentration is above c*. However, the effects of microstructure at concentrations below c* are unclear. This is especially important for practical design rules considering that c* corresponds to zero shear viscosities of 22.0 – 55.2 mPa·s for guar gum, hydroxypropyl methyl cellulose (HPMC), and λ–carrageenan, indicating that many beverages would be at usage levels below c* (Cook et al., 2003). Moreover, most studies have evaluated hydrocolloid thickened, sweetened aqueous solutions, making it difficult to develop practical design rules for more complex beverages with targeted applications that may include combinations of proteins, fats, and hydrocolloids.

As a result, the focus of the present study was to develop a system based on milk protein concentrate (MPC) that would more closely resemble the complex beverages targeted for meal replacement or specific health-related applications. The objectives of this study were to assess the effects of viscosity on sweetness perception in solutions thickened with sodium carboxymethyl cellulose (CMC) at concentrations below and above c*, and to determine if the same trends applied to a more complex beverage containing CMC at similar concentrations with the addition of MPC. Additionally, time intensity of sweet taste was evaluated to determine if taste suppression could be explained by an alteration of the temporal profile.

3. MATERIALS AND METHODS

3.1. Model system development

Food grade medium viscosity sodium carboxymethyl cellulose (CMC) with a reported 250,000 g mol^{-1} average molecular weight and 0.7 degree of substitution was purchased from Sigma Aldrich (St. Louis, MO, USA). The CMC was hydrated in deionized water by magnetic agitation at 300 rpm for 2 h. Stock solutions were stored 12 h at 4 ˚C, and the pH was adjusted to 6.9 ± 0.05 using 1 M NaOH or HCl before use. For sensory analysis, the CMC was dry mixed with sucrose purchased from a local grocery immediately prior to hydration. Milk protein concentrate (MPC) was provided by Fonterra (Auckland, NZ). Stock MPC solutions were prepared by dispersing powder into deionized water at 600 rpm for 60 s, then in a 55 ˚C water bath for 120 min. Solutions were stirred using an overhead mixer (Heidolph RZR, Schwabach, Germany) with attached pivoting blade impeller (60 mm diam.) at 250 rpm to ensure complete hydration. The final composition of the model system containing MPC was 6.0% w/w sucrose, 3.0% w/w protein, 1.8% w/w fat, and 1.5% w/w lactose. Samples were stored no longer than 3 days prior to instrumental or sensory analysis. No macroscopic phase separation was observed during this time.

3.2. Rheological flow profiles of model system

Rheological flow profiles of CMC and MPC + CMC samples were determined at 20 ± 0.05 ˚C using an Anton Paar MCR 302 rheometer (Graz, Austria). Low viscosity samples were measured using double gap cup and bob geometry (cup OD = 27.6 mm, bob OD = 26.6 mm, 500 μm wall gap), whereas high viscosity samples were measured with 1˚ cone and plate geometry (55 mm diam., 100 μm gap). Samples were sheared at 10 s^{-1} for 10 s and quiescently held for another 10 s immediately before data collection. Logarithmic shear rate sweeps were performed from 0.08 to 1000 s^{-1}. Data at or below the reported torque sensitivity (1 μN m) of the instrument were omitted.

To determine critical overlap concentration of CMC solutions, shear rate sweeps were performed from 0.1 to 3000 s^{-1} at 20 ± 0.05 ˚C. Double gap cup and bob geometry was used for low viscosity samples and the cone and plate geometry for high viscosity samples. Data with torque values below 1 μN m were omitted from the model. Zero shear viscosity (η_0) was determined using the Cross model (Eq. 1), where η_∞ is the infinite shear viscosity, $\dot{\gamma}$ is the

shear rate, and *a* and *p* are model parameters representing time and rate constants, respectively (Steffe, 1996).

$$\eta = \eta_\infty + \frac{\eta_0 - \eta_\infty}{1 + a\dot{\gamma}^p} \quad (1)$$

Intrinsic viscosity [η] was determined at 40 ± 0.2 ˚C using a Cannon-Fenske (Cannon Instruments, State College, PA, USA) capillary viscometer per the method described by Wagoner & Foegeding (2017). Solutions of CMC were prepared and diluted with 50 mM NaCl in order to account for the polyelectrolyte behavior of CMC (Pals & Hermans, 1952). Two measurements were performed 10 min apart and averaged for each datum. Specific viscosity (η_{sp}) and relative viscosity (η_{rel}) were calculated using Eq. 2 and Eq. 3, respectively, where *t* is the efflux time of the sample solution and t_0 is the efflux time of the solvent.

$$\eta_{sp} = \frac{t - t_0}{t_0} \quad (2)$$

$$\eta_{rel} = \frac{t}{t_0} \quad (3)$$

Plots of η_{sp}/c (i.e., reduced viscosity, η_{red}) and η_{rel}/c (i.e., inherent viscosity, η_{inh}) as a function of CMC concentration were used to determine [η] via extrapolation of best-fit line to zero CMC concentration per the Huggins (Eq. 3) and Kraemer (Eq. 5) equations, where k_H is the Huggins constant for a given solvent, k_K is the Kramer constant for a given solvent, *c* is polymer concentration (Tanford, 1961). This technique is valid for solutions with η_{rel} values up to ~ 2.

$$\frac{\eta_{sp}}{c} = [\eta] + k_H[\eta]^2 c \quad (4)$$

$$\frac{\ln(\eta_{rel})}{c} = [\eta]c + k_K[\eta]^2 c \quad (5)$$

3.3. Confocal Laser Scanning Microscopy

Microstructural features of fluids were investigated using a Zeiss LSM 800 confocal laser scanning microscope (Carl Zeiss AG; Oberkochen, Germany). Samples were stained with a mixture of Nile Red (0.5% w/w in polyethylene glycol), Fast Green FCF (0.2% w/w in polyethylene glycol), and Fluorescent Brightener 28 (0.1% w/w in polyethylene glycol) to visualize dispersed fat, protein, and carboxymethyl cellulose, respectively. Samples were combined with stains at a 1:50 volume ratio of stain to sample and mixed in Eppendorf tubes for 10 min. The objective lens used were 40X and 63X oil immersion lens and a 1.8X digital zoom was applied in the software (Zeiss Zen Version 2.3). Excitation wavelengths were 488 nm, 640 nm, and 405 nm. Digital image files were obtained at a resolution of 1024 by 1024 pixels. Images in the *xy* plane were obtained at a *z*-depth of at least 5 μm to avoid artifacts from the cover glass slip.

3.4. Sensory descriptive analysis of taste and texture attributes

All sensory testing was performed at North Carolina State University in compliance with the North Carolina State University Institutional Review Board for Human Subjects. Descriptive sensory analysis of texture attributes and sweet taste was performed using a trained panel (n = 7; 1 male, 6 females ages 24–69 years) of members of an existing descriptive analysis panel in the Department of Food, Bioprocessing, and Nutrition Sciences at North Carolina State University. This panel had been trained in the Spectrum™ method of descriptive analysis for generation of qualitative and quantitative data (Drake & Civille, 2003; Meilgaard, Civille, & Carr, 2007). Each panelist had 100+ hours experience on descriptive analysis of taste and texture. Attributes shown in Table 3.1 were based on established lexica for fluids (Campbell, Daubert, Drake, & Foegeding, 2016; Pollen, Daubert, Prabhasankar, Drake, & Gumpertz, 2004). All attributes were reviewed for appropriateness by the panel during a training session using samples representative of the experimental treatments and calibration solutions for sweet taste. A 15-point universal scale was used where 0 represented no intensity and 15 represented maximum intensity. Samples were presented at 20 ˚C in 59 mL plastic soufflé cups coded with random three-digit numbers. Each panelist evaluated each sample in triplicate over the course of two sessions on two

separate days. Distilled, deionized water and unsalted crackers were used as palate cleansers between samples.

Table 3.1. Sensory lexicon and reference solutions used for descriptive sensory analysis.

Term	Definition	References (0–15 point scale)
Slurp viscosity	Force required to initiate flow from spoon	Water: 1; heavy cream: 3
Viscosity in mouth	Sample thickness in mouth	Water: 1; heavy cream: 3
Adhesiveness	Degree to which the fluid sticks to oral surfaces	None
Mouth coating	Number of particles or film remaining in the mouth after swallowing	None
Sweet taste	Basic taste	5% sucrose: 5; 7% sucrose: 7; 9% sucrose: 9

3.5. Time intensity of sweet taste

All panelists (n = 7; 1 male, 6 females ages 24–69) had at least 100 hours of descriptive analysis training. Three 1 h training sessions were used to familiarize panelists with similar samples and the time intensity method. All samples were served as 20 mL portions at 20 ˚C in a lidded 59 mL soufflé cup assigned a random three-digit code. Panelists were calibrated using 4, 5, 6, and 7% w/w sucrose solutions prior to evaluating each sample. Panelists were instructed to place the entire sample in the mouth and the signal to expectorate appeared after 12 s. Sweetness intensity was evaluated for the 12 s prior to expectoration and an additional 90 s using a 15–point Spectrum™ scale. Data were collected via a sliding scale on iPads using Compusense Cloud (Compusense, Guelph, Canada). A 5 min rest period was enforced between each sample, and panelists were instructed to use distilled, deionized water and unsalted crackers as palate cleansers. Each panelist evaluated each sample four times over the course of two sessions.

Table 3.2. Definitions of sensory parameters used in time intensity analysis

Attribute	Definition
T_{max} (s)	Time to reach maximum intensity
I_{max}	Maximum perceived intensity at any time
Initial delay (s)	Time elapsed prior to onset of sweet taste
Duration (s)	Total duration of perceived intensity
Plateau length (s)	Time difference between reaching maximum and the beginning of decline
Increasing angle (˚)	Rate of increase (linear regression) from onset to maximum intensity
Decreasing angle (˚)	Rate of decrease (linear regression) from initial declining point to baseline
Area under the curve	Total area under time intensity curve

3.6. Statistical analysis

Data are presented as the arithmetic mean of three independent replications ± standard deviation unless otherwise indicated. Statistical analysis was performed using JMP Pro software version 13.0 (SAS, Cary, NC, USA). Treatment effects for descriptive analysis were determined using one-way analysis of variance (ANOVA) followed by Tukey's HSD as a *post hoc* multiple comparisons test to separate means where necessary. Experiment wise error was set at $\alpha = 0.05$ for each response. Statistical analysis of time intensity of sweet taste was performed using two-way ANOVA accounting for panelist × treatment interactions followed by a *post hoc* means separation using Tukey's HSD ($\alpha = 0.05$). Time intensity attributes shown in Table 2 were calculated within Compusense Cloud individually for each replication by each panelist. Principal component analysis (PCA) was used to visually represent how time intensity parameters, descriptive analysis attributes and measures of viscosity differentiated the samples. Analysis was performed using XLSTAT v18.01 (Addinsoft, New York, NY, USA).

4. RESULTS

4.1. Determination of intrinsic viscosity

Intrinsic viscosity, [η], was determined in order to calculate the critical overlap concentration, c*, of the medium viscosity carboxymethyl cellulose, CMC, used in the study. Intrinsic viscosity was experimentally determined to be 5.86 ± 0.08 dL/g (Fig. 3.1).

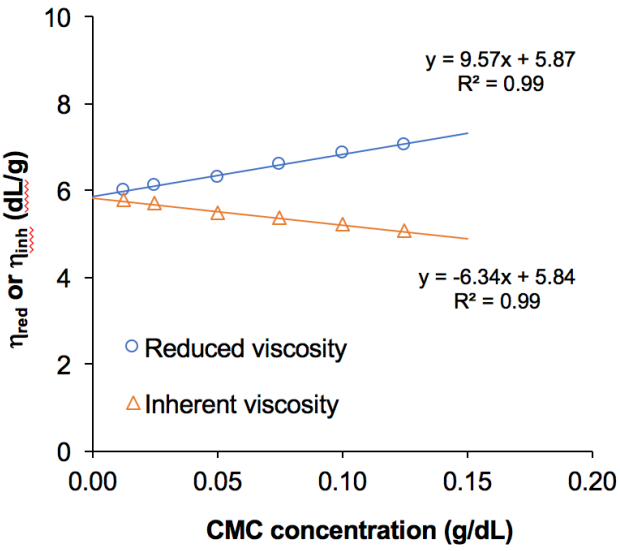

Figure 3.1. Reduced and inherent viscosity of carboxymethyl cellulose as a function of concentration.

4.2. Determination of CMC critical overlap concentration, c*

Determination of critical overlap concentration requires a shear-rate independent viscosity, so the zero-shear viscosity, η_0, of CMC solutions was calculated using shear rate sweep data fit to the Cross model. An abrupt transition in η_0 with increasing c[η] was not observed, but rather a gradual increase in η_0 with increasing c[η], likely due to polydispersity (Fig. 3.2). The c* determined from intersection of the two best fit lines was c[η] = 3.93. Although [η] is proportional to molecular weight, c[η] is normalized by concentration and hydrocolloids align to a master curve at c[η] = 4, which agrees with our experimental data

(Morris, Cutler, Ross-Murphy, Rees, & Price, 1981). The c* corresponds to 0.67 % w/w CMC and a η_0 of 22 mPa·s, which is similar to reported η_0 of 22 – 55 mPa·s for guar gum, HPMC, and λ–carrageenan (Cook et al., 2003).

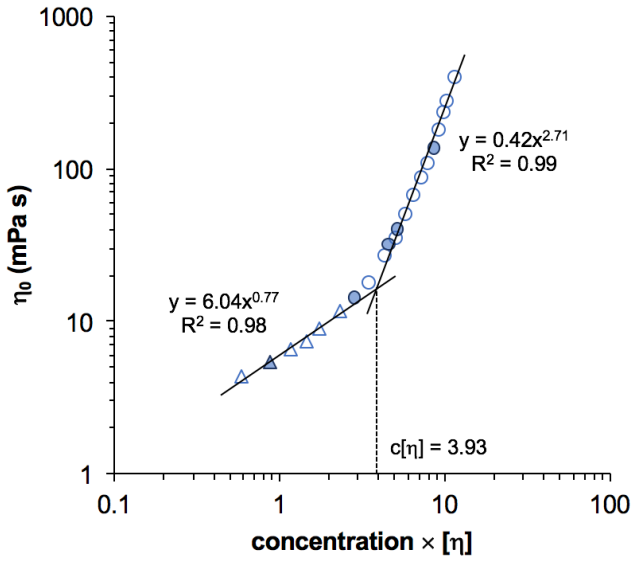

Figure 3.2. Determination of carboxymethyl cellulose critical overlap concentration. Zero shear viscosity, η_0, was determined using double gap cylinder (△) and cone and plate geometries (○). Intrinsic viscosity, [η], was experimentally determined to be 5.86 dL/g. Filled symbols represent samples selected for subsequent experiments.

4.3. Flow profiles of CMC solutions with sucrose

Based on c*, five CMC samples were initially selected (0.15, 0.5, 0.8, 0.9, and 1.5% w/w) in order to target discrete concentrations relative to c*. A sucrose level of 6.0% was selected as a common usage level in beverages. All samples exhibited a low shear Newtonian plateau and shear thinning behavior (Fig. 3.3). The onset shear rate for the shear thinning region decreased with increasing CMC concentration. The η_0 were significantly distinct ($p <$ 0.05) across all CMC concentrations: 5.5 mPa·s, 14.0, 31.0, 40.5, and 133.3 mPa·s for 0.15, 0.5, 0.8 0.9 and 1.5% w/w CMC, respectively.

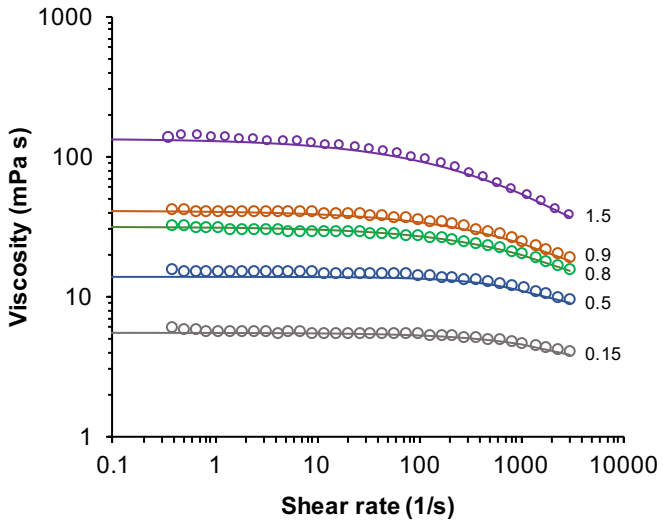

Figure 3.3. Shear rate dependent viscosity of carboxymethyl cellulose solutions. Samples contain 6.0% w/w sucrose. Experimental data are represented by symbols, and solid lines represent data fit to the Cross Model. Numbers represent the mass percentage of carboxymethyl cellulose.

4.4. Descriptive analysis of CMC solutions with sucrose

Six attributes were selected for descriptive sensory analysis, including sweet taste, slurp viscosity, in-mouth viscosity, adhesiveness, mouth coating, and in-mouth smoothness. However, no samples exhibited any degree of phase separation or grittiness and thus did not differ in in-mouth smoothness (therefore data have been omitted). Descriptive analysis of CMC solutions with 6.0% w/w sucrose is shown in Table 3.3. A significant reduction in sweet taste was observed at 1.5% w/w CMC; a concentration above c* that corresponded to a η_0 of 133 mPa·s. In contrast, 0.8 and 0.9 % w/w CMC were slightly above but close to c*, yet no sensory difference was observed. In terms of textural attributes, samples were well-differentiated in terms of slurp viscosity, in mouth viscosity, adhesiveness and mouth coating after expectoration, all of which increased with CMC concentration. Increased slurp and in-mouth viscosity strongly agree with instrumentally measured viscosity. The samples with 0.8 and 0.9% w/w CMC were very similar across all attributes, except for 0.9% CMC being significantly more mouth coating.

Table 3.3. Descriptive analysis of carboxymethyl cellulose (CMC) solutions with sucrose.

CMC (%)	Sweet taste	Slurp viscosity	In-mouth viscosity	Adhesiveness	Mouth coating
0.15	6.0 ± 0.1a	1.6 ± 0.2d	1.9 ± 0.2d	0.8 ± 0.2d	1.3 ± 0.2e
0.50	6.0 ± 0.1a	2.0 ± 0.2c	2.3 ± 0.2c	1.2 ± 0.4c	1.7 ± 0.2d
0.80	6.0 ± 0.1a	2.8 ± 0.3b	3.2 ± 0.3b	2.1 ± 0.4b	2.3 ± 0.2c
0.90	6.0 ± 0.1a	3.0 ± 0.3b	3.3 ± 0.3b	2.2 ± 0.2b	2.7 ± 0.2b
1.50	5.8 ± 0.1b	5.9 ± 0.4a	6.3 ± 0.4a	3.6 ± 0.3a	3.8 ± 0.4a

Samples contain 6.0 % w/w sucrose. Values scaled on 15-point Spectrum scale. Letters indicate significant differences within each column as determined using one-way ANOVA followed by Tukey's HSD *post hoc* test.

4.5. Flow profile of MPC, CMC, and sucrose solutions

Flow profiles of MPC and CMC solutions with 6.0% w/w sucrose are shown in Fig. 3.4. The MPC without added CMC is nearly Newtonian with viscosity near 2–3 mPa·s across most shear rates. Contrary to the CMC flow profile, the mixture of CMC and MPC did not exhibit a low-shear Newtonian plateau across the measured shear rates. The binary mixture of CMC and MPC exhibited viscosity greater than the sum of the individual ingredients, likely indicating interactions between molecules. The mixture of MPC and CMC also did not exhibit a η_∞ plateau across the measured shear rates.

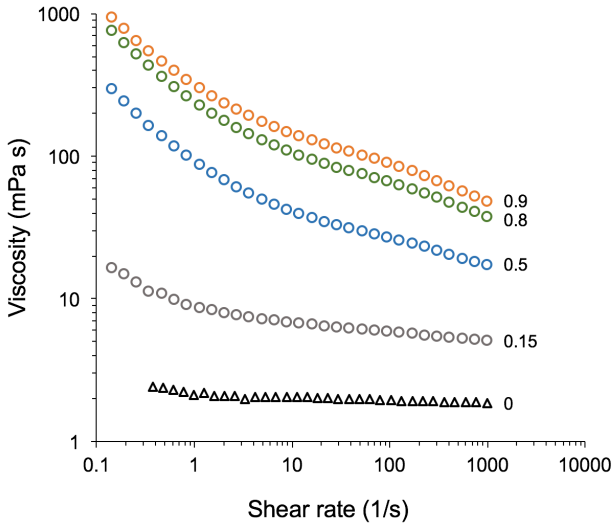

Figure 3.4. Shear rate dependent viscosity of MPC and CMC solutions with 6.0% sucrose. Solutions were matched at 3.0% w/w protein, and numbers next to data indicate CMC mass percentage. Numbers represent the mass percentage of carboxymethyl cellulose.

4.6. Confocal laser scanning microscopy of MPC, CMC, and sucrose solutions

Confocal micrographs of MPC and CMC solutions are shown at two magnification levels in Fig. 3.5. The MPC without added CMC formed a relatively homogenous dispersion of fat droplets suspended in a proteinaceous aqueous phase. The microstructure was similar upon addition of 0.15% CMC and the blue background suggests both protein and CMC comprise the aqueous phase. At 0.5% CMC and above, micro-phase separation was observed likely due to interactions among proteins and CMC. The higher magnifications show protein-CMC aggregates with smaller amounts that appear to be predominately protein or CMC.

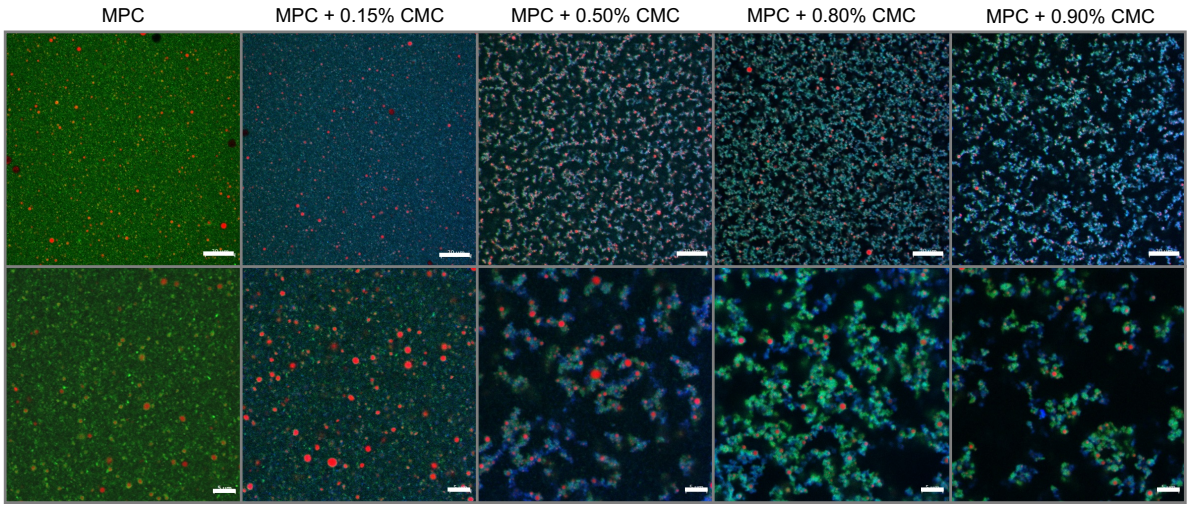

Figure 3.5. Confocal micrographs of milk protein concentrate model system with different carboxymethyl cellulose concentrations. Scale bars represent 20 μm and 5 μm in the top and bottom rows, respectively. Protein, fat, and cellulose phases are depicted by green, red, and blue colors, respectively.

4.7. Descriptive analysis of MPC, CMC, and sucrose solutions

Descriptive sensory analysis of MPC and CMC solutions with 6.0% w/w sucrose is shown in Table 3.4. The 1.5% w/w CMC concentration formed a semisolid when mixed with MPC and was thus excluded from the remainder of the study. Sweet taste was slightly higher than CMC alone at equivalent CMC concentrations, which is likely attributed to residual lactose in the MPC powder. Unlike CMC alone, MPC + CMC samples trended towards increased sweet taste with increasing CMC, with a significant increase at 0.90% CMC. MPC + CMC samples were well differentiated with significant increases in slurp viscosity, in-mouth viscosity, adhesiveness and mouth coating at each CMC level, similar to what was observed with the CMC samples. Compared to CMC alone, all texture attributes were higher for the MPC and CMC mixture. Increases in viscosity matched rheological properties, where combinations of MPC and CMC were more viscous. Mouth coating is likely due to addition of fat, although both casein and β–lactoglobulin have been shown to be mucoadhesive to porcine tissue *in vitro* (Withers et al. 2013).

Table 3.4. Descriptive analysis of milk protein concentrate model system with variable carboxymethyl cellulose and 6.0% sucrose.

CMC (%)	Sweet taste	Slurp viscosity	In-mouth viscosity	Adhesiveness	Mouth coating
0.15	6.2 ± 0.1c	2.1 ± 0.0d	1.9 ± 0.1d	1.0 ± 0.2d	1.7 ± 0.1d
0.50	6.2 ± 0.0bc	2.8 ± 0.1c	2.8 ± 0.1c	2.1 ± 0.0c	2.8 ± 0.0c
0.80	6.3 ± 0.1b	3.3 ± 0.1b	3.4 ± 0.1b	2.9 ± 0.1b	3.6 ± 0.1b
0.90	6.5 ± 0.1a	3.9 ± 0.1a	4.0 ± 0.2a	3.5 ± 0.1a	4.2 ± 0.2a

Values are scaled on a 15-point Spectrum™ scale. Letters indicate significant differences within each column as determined using one-way ANOVA followed by Tukey's HSD *post hoc* test. The 1.5% w/w CMC treatment formed a phase-separated semisolid and was omitted from subsequent tests.

4.8. Time Intensity of sweet taste

Time intensity of sweet taste profiles are shown in Fig. 3.6. Despite disparate viscosities, the profiles were very similar, with no apparent differences among CMC concentrations. It is important to note that this approach was time-averaged across all panelists, which disregards the presence of unique panelist "fingerprints" for all samples (Lawless & Heymann, 2010). The cause of unique panelist profiles is unknown but is thought to relate to oral processing efficiency or physiological differences such as salivary flow rate and receptor density. As a result, time intensity profiles are also displayed as panelist averages (Fig. 3.7). This representation underscores the unique panelists profiles of sweet taste, where individual panelist T_{max} values averaged across all CMC levels ranged from 6.7 s to 33.5 s. From the individual panelist replicates, time intensity attributes were calculated and are presented in Table 3.5. Even accounting for variability among panelists, time intensity attributes were still very similar. Only plateau length and increasing onset angle showed a significant effect ($p < 0.05$) of CMC concentration (and only between 0.15 and 0.9% w/w CMC), both decreasing inversely with CMC concentration.

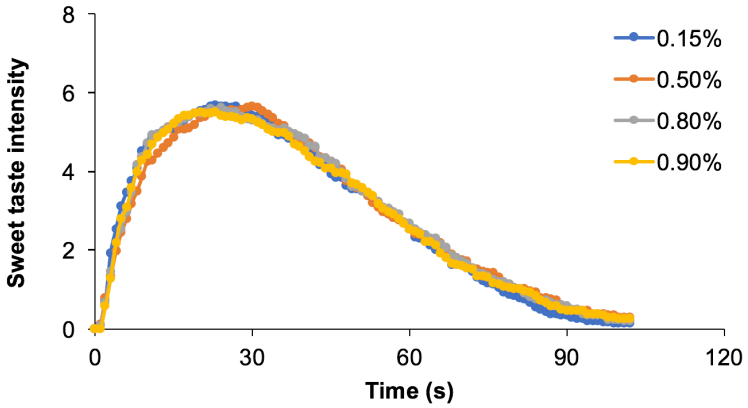

Figure 3.6. Sweet taste time intensity profiles of MPC and 6.0% w/w sucrose solutions with variable amounts of CMC. Profiles represent time-averaged data across 7 panelists and 4 independent replications.

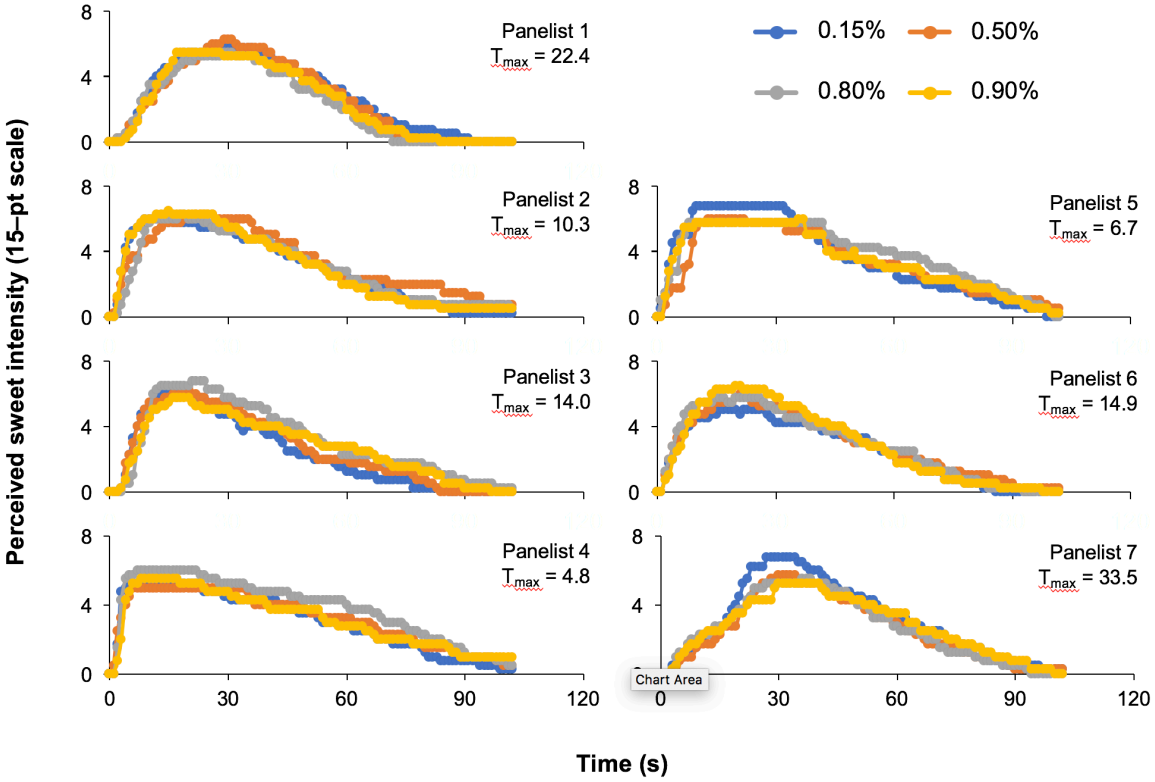

Figure 3.7. Sweet taste time intensity profiles of MPC and 6.0% w/w sucrose solutions with variable amounts of CMC represented across individual panelists. Data represents the mean of four independent replications. Time to maximum sweetness (T_{max}) was averaged across all CMC concentrations to illustrate differences among panelists.

Table 3.5. Time intensity of sweet taste parameters for milk protein concentrate with variable carboxymethyl cellulose.

CMC (%)	T_{max} (s)	I_{max}	Delay (s)	Duration (s)	Plateau Length (s)	Increasing Angle (°)	Decreasing Angle (°)	AUC
0.15	13.6	5.9	2.6	80.8	18.6a	35.8a	5.8	286
0.5	17.0	6.0	2.6	81.7	15.8ab	25.8b	6.1	292
0.8	20.3	6.1	2.7	81.9	15.9ab	28.7b	8.2	304
0.9	15.7	6.1	2.9	82.0	14.5b	30.4b	5.5	295

Letters indicate significant differences within each column as determined using one-way ANOVA followed by Tukey's HSD post hoc test. T_{max} represents time to maximum intensity, I_{max} represents maximum intensity, and AUC represents total area under time intensity curve. Parameter definitions are given in Table 2.

4.9. Principal components analysis of MPC and CMC samples

Principal component analysis of MPC and CMC samples representing time intensity attributes, descriptive sensory analysis, and viscosity at shear rates of 0.1 and 1000 s^{-1} is shown in Fig. 3.8. Factors one and two explain 93.97% of the sample variability. Samples with 0.8 and 0.9% w/w CMC load positively on F2 and were characterized by longer sweet duration, mouth coating, adhesiveness, in mouth and slurp viscosity, and measured viscosity at 0.1 and 1000 s^{-1}. A second grouping of attributes loaded positively on F1 was composed of time intensity attributes T_{max}, I_{max}, AUC, and decreasing angle.

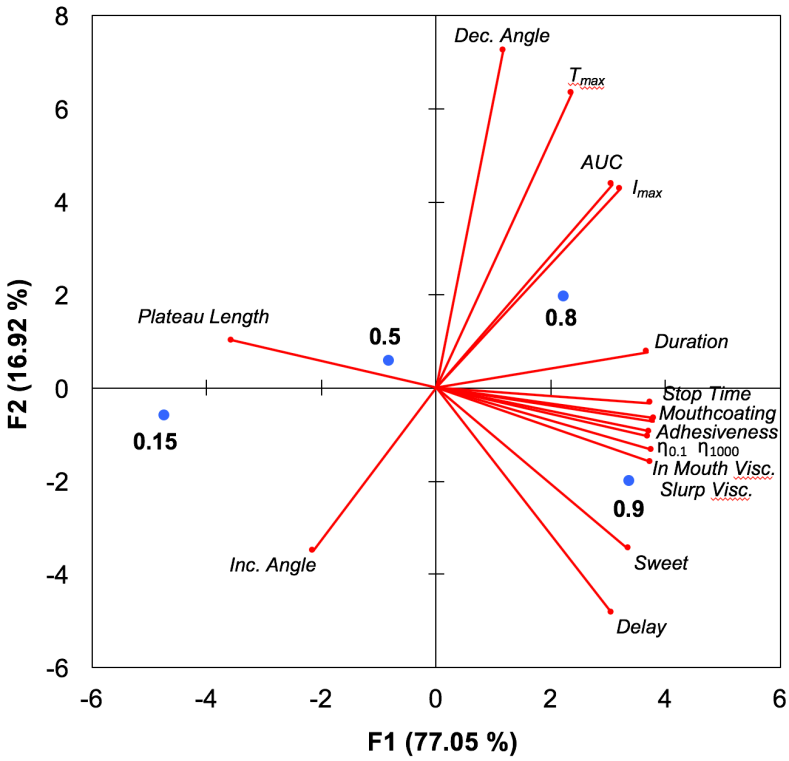

Figure 3.8. Principle component analysis (PCA) biplot (F1 and F2) of descriptive analysis, time intensity, and rheology of MPC model system with variable CMC concentration. Samples are represented by circles labeled with the corresponding CMC mass concentration, and vectors represent attributes. Abbreviations represent: area under the time intensity curve (AUC), maximum perceived intensity (I_{max}), time to reach maximum intensity (T_{max}), and viscosity at 0.1 s^{-1} ($\eta_{0.1}$) and 1000 s^{-1} (η_{1000}).

5. DISCUSSION

Beverages can be complex and contain combinations of polysaccharides, proteins, small water-soluble molecules, and fat. These components can have a profound effect on flavor, taste, and texture perception. Understanding how these components alter food structure and, consequently, taste perception is very beneficial to the food industry, and has been the subject of repeated investigations from as early as the 1960s. Viscosity-taste interactions are also important from a health perspective, where increased viscosity has been associated with positive health outcomes such as and reduced hunger and *ad libitum* intake (de Wijk et al., 2008; Mars et al., 2009). However, despite long standing interest in viscosity-

taste interactions, a comprehensive understanding of the role of food structure and associated properties, such as viscosity, in taste perception is still unclear.

Based on conclusions of early studies in the 1970s, it has been widely reported that increasing fluid viscosity through that addition of hydrocolloids suppresses taste on the basis of restricted diffusion (Arabie & Moskowitz, 1971; Pangborn et al., 1973). More recent studies have indicated that taste suppression may not occur until hydrocolloid concentrations increase beyond the critical overlap concentration, c* (Baines & Morris, 1987; Cook et al., 2002; Han et al., 2014; Hollowood et al., 2002). The c* refers to the concentration at which hydrodynamic domains of adjacent polymers overlap and result in a marked increase in viscosity. At concentrations above c*, taste suppression is thought to be a result of inefficient mixing between the polymeric solution and saliva (Baines & Morris, 1987). Based on tastant mass transfer models, this is thought to reduce the flux of tastants to receptors, which is one major driver of overall perceived intensity (Cook et al., 2003; Kokini, 1987; Pfeiffer et al., 2000). The phenomenon of critical overlap taste suppression has been reported for CMC, hydroxypropyl methyl cellulose (HPMC), guar gum, and locust bean gum (Baines & Morris, 1987; Cook et al., 2002; Ferry et al., 2006; Han et al., 2014; Koliandris, Lee, Ferry, Hill, & Mitchell, 2008; Koliandris et al., 2010).

In the present study, CMC was used to increase viscosity on account of being colorless and flavorless. The CMC was characterized to determine c* and a reduction in sweet taste with increasing CMC concentration was observed only at one concentration (1.5% CMC) well above c*. The CMC concentrations near but just slightly above c* (0.8 and 0.9% CMC) were not significantly different from those below c* (0.15 and 0.5% CMC). Considering the numerous reports of decreased taste perception above c*, the observed decrease in sweet taste at 1.5% CMC is not surprising. However, in terms of rational food design, it should be noted that this concentration of CMC had a η_0 of 133 mPa·s, agreeing with a study on taste perception in sauces thickened with HPMC, where suppression did not occur until a viscosity near 280 mPa·s (Ferry et al., 2006). This is in contrast to the conclusion of Pangborn et al. (1973) that viscosity greater than 16 mPa·s decreased sucrose sweetness.

The other consideration that informed the experimental design of this study is that previous evaluations of texture-taste interactions of fluids most often used water as a

medium. The simplicity of this approach is important for limiting potential confounding variables in mechanistic investigations; however, most beverages contain other ingredients – especially considering the growing consumer demand for protein. It is currently unknown if the taste suppression that occurs in aqueous systems applies to fluids with more complex microstructures and additional ingredients, and more importantly, at what hydrocolloid concentrations taste attenuation may become a detectable difference. Thus, an MPC fluid system thickened with CMC was developed in this study where the inclusion of fat and protein are more representative of complex beverages on the market. An MPC derived from whole milk was used to approximate the macromolecular profile of milk (3.0% protein and 1.8% fat) with lower lactose (1.5%) to control the sweetness level with sucrose (6.0%). From a microstructure perspective, the use of this MPC allows for a complex system with colloidal components spanning multiple length scales.

When dispersed in water, only 1.5% CMC suppressed sweet taste. However, this level was not feasible in combination with MPC due to the formation of a semisolid. At 0.8 and 0.9% CMC – both still slightly above c* – sweet taste was not suppressed and instead significantly ($p < 0.05$) increased. There is precedence for this somewhat surprising increase in sweet taste with viscosity, with similar reports for CMC and propylene glycol alginate in fermented whey beverages (Gallardo-Escamilla, Kelly, & Delahunty, 2007), and for saccharin solutions thickened with CMC (Pangborn et al., 1973). However, despite being *statistically* significant, an increase of 0.2 on a 15-point scale may not be *practically* significant or detectable for the average consumer. Instead, it highlights that in this type of system, viscosity can be adjusted over a large range without concerns for taste suppression.

Based on mass transfer models of sweet taste, it is hypothesized that the increased viscosity and high degree of mucoadhesion allowed tastant molecules to remain in close proximity to receptors (Aubert et al., 2016; He et al., 2016; Koliandris et al., 2010). The concept of mucoadhesion is only beginning to be addressed in food science, despite being leveraged by the pharmaceutical industry for decades in order to maximize contact with mucosa for enhanced drug delivery (Cook, Bull, Methven, Parker, & Khutoryanskiy, 2017; Smart, 2005). Moreover, as an anionic polysaccharide CMC is an excellent mucoadhesive and reported to be more mucoadhesive than non-ionic HPMC and guar gum (Nafee, Ismail, Boraie, & Mortada, 2004). Sensory data from this study corroborates mucoadhesion, where

mouth coating and oral adhesiveness increased with CMC usage even in the absence of milk proteins. The addition of MPC further increased these attributes. It should be noted that in addition to interactions between ingredients and mucosal surfaces, mucoadhesion is also a function of viscosity alone, making it difficult to isolate individual contributions to oral adhesion perception.

Maximum perceived sweetness also depends on the onset and temporal progression of sweet taste (Bayarri, Rivas, Izquierdo, & Costell, 2007). This may indicate that the overall perceived intensity during descriptive analysis could be explained by an altered temporal profile (i.e., change in flux of tastants to receptors). However, after measuring the time intensity of sweet taste, no significant differences ($p > 0.05$) were observed in time-averaged profiles across all panelists. This is likely due to the variation in individual panelist profiles. These unique panelist "fingerprints" may indicate physiological differences that alter the progression of taste; however, this could also highlight the role of oral processing (i.e., the summation of tongue and jaw movements) in modifying taste perception. We then accounted for panelist variability (two-way ANOVA with viscosity by panelist interaction) and observed a significant viscosity effect only between 0.15% and 0.90% CMC for increasing slope and plateau length. Increasing slope at 0.15% CMC is indicative of a more rapid onset of maximum sweet taste, which may also explain the longer plateau length. Multivariate analysis also indicated that CMC concentration tended to correlate with slightly delayed onset and longer total duration. Thus, it is possible that a viscosity effect is present but even larger differences in viscosity would be needed to significantly alter the time intensity parameters.

The presence of multimodal interactions may have also played a role in the results of this study. Overall perception is the interaction of olfactory, gustatory, and somatosensory tactile sensations, underscoring the role of cognition in total taste perception (Calvert, Brammer, & Iversen, 1998; Rolls, 2005). This makes it particularly challenging to study the effects of texture on taste or flavor perception – even in simple polymer and water solutions – where modifying viscosity also modifies flavor release and mouthfeel attributes (Cook et al., 2003; Ferry et al., 2006). This also suggests a strong cognitive component, as more viscous samples are reported to exhibit less intense flavor despite the measured concentration of volatile compounds in the nasal cavity being unchanged (Cook, Hollowood, Linforth, &

Taylor, 2005; Hollowood et al., 2002). Additionally, changes in fat level influence taste perception (Malone, Appelqvist, & Norton, 2003). The discrepancy between the results of this study and previous studies in aqueous solutions could be due to the presence of fat.

Despite confirmation that polymer concentrations above c* suppress taste, the mechanism of this effect is still unknown. One could argue that a high degree of polymer entanglement effectively generates a transient structure where tastant molecules need to be released in order to mix with saliva and reach receptors. This effect may be dependent on the tastant concentration. Tournier et al. only reported an effect of custard viscosity on taste perception at 7.0% sucrose; at 3.5% sucrose (and using lactose monohydrate to keep total solids constant without increasing sweetness) there was no effect of texture on taste (Tournier et al., 2009). Thus, at sweetness equivalencies other than the 6.0% sucrose used in the present study these trends may change.

To our knowledge, this is the first study to build on the structural aspects of the c* taste suppression principles developed for aqueous polymer solutions by applying them to a complex beverage. As with critical overlap in polymer solutions, the differences in perceived sweet taste of the MPC-CMC beverage could be an effect of microstructure. Although the aqueous CMC system cannot be visualized using confocal microscopy, structural differences would be expected between the microphase separated dairy system and the polymer system. Therefore, there may be an effect of microstructure even in a fluid system where we normally wouldn't think of structure existing. One could argue that polymer entanglement forms a sort of structure that is present in the polymer only system, and the additional of other ingredients (i.e., generating a more complex microstructure) modifies or removes the entangled structure. Ferry et al. hypothesized that the microstructural differences between granular (i.e., modified starch that maintained granular integrity) and polymeric structures (i.e., HPMC) explained why starch suppressed sweet taste less than HPMC (Ferry et al., 2006). Microstructural differences were observed in the present study, where solutions transitioned from relatively homogenous to micro-phase separated at 0.5% CMC and above. Phase separation may have been associative due to electrostatic interactions between CMC and casein at near neutral pH, or due to depletion flocculation (Dickinson, 2006). If sucrose preferentially partitions to the non-polymer serum phase, then mass transfer would not be the same as a purely polymeric system. A similar effect has been reported for hydrocolloid gels, where greater serum release

during oral processing lead to increased sweet taste perception (Sala, Stieger, & van de Velde, 2010). This underscores the importance of considering structure in taste perception even in fluid systems.

6. CONCLUSIONS

The results of this study indicate that perceived sweet taste is suppressed in CMC solutions at concentrations well above c*, whereas no effect was observed slightly above or well below c*. However, when dispersed in combination with MPC, the same CMC concentrations did not suppress sweetness and caused a significant increase at 0.90% CMC. This may indicate that the widely-reported taste suppression in polymer solutions above c* may not apply to more complex beverages containing additional ingredients and producing a more complex microstructure. The change in perceived sweet taste with increasing viscosity in solutions containing CMC and MPC could not be explained on account of differences in time intensity, suggesting that a delayed temporality does not account for the slight increase in sweet taste with increasing CMC.

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CHAPTER 4. FOOD TEXTURE AND SWEETENER TYPE MODIFY SWEETNESS PERCEPTION IN WHEY PROTEIN-BASED MODEL FOODS

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Food texture and sweetener type modify sweetness perception in whey protein-based model foods

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Abstract

In an effort to formulate healthier food products, sugar is often reduced or replaced with artificial or natural non-nutritive sweeteners. However, the removal of sucrose often changes food texture, and non-nutritive sweeteners inherently exhibit differences in sweetness quality, onset, and overall intensity. Therefore, the objectives of this study were to evaluate the effects of texture on sweetness dose-response profiles of whey protein solutions sweetened with sucrose, sucralose, stevia, or monk fruit extracts, and to assess temporal progression of dominant attributes. Whey protein-based model foods with different textural properties were generated without modifying composition by varying heating time. Dose-response power functions and iso-sweet equivalencies were determined using magnitude estimation scaling. Temporal profiles of dominant texture, taste, and flavor attributes were evaluated using temporal dominance of sensations (TDS). A significant texture-sweetener interaction effect was observed ($p < .05$), where more viscous or semisolid textures required greater amounts of sweetener for iso-sweetness. Moreover, the dose-response slope decreased with increasing thickness for sucralose, stevia, and monk fruit-sweetened textures. Results of TDS indicated that increased thickness prolonged the dominance of several attributes. This study shows that sugar reduction strategies are not universal; food matrix, sweetener type, and usage level need to be considered when attempting to modify texture or reduce sucrose.

Practical applications

Sucrose reduction, use of nonnutritive sweeteners, and texture modification are all ways in which food manufacturers can increase the healthful quality of the food supply. However, these types of modifications can alter the overall sensory properties of a food. Texture-taste interactions are not currently well understood, and knowledge in this area is important for designing ways to modify texture or reduce sucrose for health and enjoyment. The results of this study provide insight into texture-taste interactions, indicating that sugar reduction or replacement strategies are not one size fits all and are likely product and sweetener-specific.

1 | INTRODUCTION

The association of negative health effects with excess sucrose consumption have been widely reported. As a result, consumers and food manufacturers are increasingly turning to alternative sweeteners in commercial food products to reduce or replace sucrose (Pawar et al., 2013). While alternative sweeteners are useful in decreasing total calories, they differ in temporal intensity and often exhibit undesirable off-flavors and a lingering aftertaste (Cardoso & Bolini, 2008). Additionally, sucrose serves as a structural component in many food applications; thus, removal of sucrose can detrimentally impact food structure and perceived texture.

Research over the last several decades implies a role of texture in satiation and satiety, where thicker foods—or more generally foods that require a greater amount of oral processing—are associated with decreased ad libitum intake and lower reported hunger levels (de Wijk et al., 2008; Mars et al., 2009). Whereas increasing viscosity would be a positive modification for reducing hunger, viscosity has been reported to attenuate sweet taste (Baines & Morris, 1987; Cook et al., 2003; Ferry et al., 2006; Hollowood et al., 2002). Adding more sweetener to reach an equal level of sweetness would therefore negate the health benefits of thickening a product. Sensory perception of taste and flavor vary depending on food structure and breakdown during oral

processing, and texture-taste interactions modify not only the maximum perceived intensity but also the temporal progression of other attributes (Bayarri et al., 2007; Foster et al., 2011; Mosca et al., 2015; Wilson & Brown, 1997). Therefore, understanding the mechanisms of texture-taste interactions is critical for the design of foods for health and enjoyment.

Much of the research examining the effects of texture on sweetness perception has involved either fluids or soft solids. In fluids, increasing the viscosity is associated with taste and flavor suppression for a number of different polymer and starch systems (Baines & Morris, 1987; Cook et al., 2003; Ferry et al., 2006; Hollowood et al., 2002). In soft solid foods, the release of fluid during eating and/or fracturing into many small particles increases the flux of tastant molecules to sweet receptors and thus perceived sweet taste intensity (Knoop et al., 2013; Mosca et al., 2015; Stieger & van de Velde, 2013). However, few studies have examined sweetness perception across a broad range of consumer food textures, from liquids to semisolids. More importantly, these studies often modify composition or ingredients to generate textural differences in the sample foods, which could also play a role in modifying taste perception.

Sucrose is the most commonly used sweetener in the world and is preferred for its familiar taste and linear dose response (DuBois et al., 1991; Fujimaru et al., 2012). However, consumer trends toward calorie reduction and "no added sugar" labeling have contributed to a marked increase in the consumption of non-nutritive sweeteners (Sylvetsky & Rother, 2016). Sucralose is an artificial sweetener originally sold under the name Splenda and is reported to be 320 to 1,000 times sweeter than sucrose (Pawar et al., 2013). Since approval in 1999, sucralose has become the most commonly used non-nutritive sweetener in the food industry (Sylvetsky & Rother, 2016). Consumer demand for natural, non-nutritive sweeteners has led to the recent popularity of stevia and monk fruit extracts (Pawar et al., 2013). *Stevia rebaudiana* is a woody shrub found in many parts of the United States and South America. It is a source of a number of sweet tasting *ent*-kaurene diterpenoid glycosides, primarily dulcoside A (3–5%), rebaudioside A (20–25%), rebaudioside C (5–10%), and stevioside (50–55%) (Mizutani & Tanaka, 2002; Shibasato, 1995). As a food ingredient, stevia needs to contain at minimum 95% steviol glycosides with rebaudioside A and/or stevioside as the predominant components (FDA, 2018). The individual glycosides have disparate sweetness intensities and flavor profiles that vary according to concentration and environment (DuBois & Prakash, 2012). Collectively, they make stevia 100 to 300 times sweeter than sucrose depending on composition and usage level (Cardello et al., 1999). Stevia is stable at high temperatures and across a wide pH range (Sirshendu et al., 2013). Monk fruit extracts are prepared from the fruit of (also known as luo han guo) the flowering vine *Siraitia grosvenorii* (Pawar et al., 2013). The sweet components are cucurbitane-type triterpene glycosides, namely mogroside V as the most concentrated constituent at up to 55% by weight; mixtures of these glycosides are reported to be 250 to 425 times sweeter than sucrose (Kim & Kinghorn, 2002).

Several studies have evaluated sucrose replacement with sucralose, stevia, or monk fruit extracts in a variety of foods (Leksrisompong et al., 2012; Li et al., 2015; Morais et al., 2014; Paixão et al., 2014; Palazzo & Bolini, 2014; Reyes et al., 2017). The results of these studies indicate that replacement of sucrose alters the temporal profile of sweetness and other sensory attributes. Additionally, significant texture-taste interactions are present and thus the replacement ratios vary by food texture. Food composition also plays a role, where the addition of fat or dairy and cocoa solids alters dose-response profiles and iso-sweet concentrations (Li et al., 2015; Paixão et al., 2014). However, the specific effect of texture—without varying composition—has not been explored. Thus, there is a need to further elucidate texture-taste interactions in the context of varying texture without modifying formulation.

This study aimed to determine how sweetness perception of sucrose, sucralose, stevia, and monk fruit varied among thin fluid, thick fluid, and semisolid food structures. A whey protein isolate solution was selected as a model system because distinct structures with different perceived textures could be generated without modifying formulation by varying the total heating time (Campbell et al., 2016). Magnitude estimation scaling (MES) was used to identify sweetener concentrations required for equal perceived sweet taste intensity and slope of the dose-response power function. Temporal dominance of sensations (TDS) was then used to evaluate the temporal effects of texture and sweetener on perceived dominant attributes.

2 | MATERIALS AND METHODS

2.1 | Sample preparation

A model system consisting of three iso-caloric, macronutrient-matched foods flavored with vanilla flavor was used (Campbell et al., 2016). A commercial whey protein isolate (WPI) produced by ion exchange chromatography (93.21% dry basis protein based on a Nitrogen conversion factor of 6.38) was obtained (Agropur Cooperative, Eden Prairie, MN). Individual whey protein content determined by reverse phase HPLC consisted of 69.2% β-lactoglobulin, 30.8% α-lactalbumin, and negligible bovine serum albumin or glycomacropeptide (Wagoner et al., 2015). Elemental content of WPI was determined by inductively coupled plasma atomic emission spectroscopy and contained 0.06% P, 0.77% K, 0.08% Ca, 0.04% Mg, 1.31% S, and 0.69% Na on a weight basis. Granulated cane sugar (Domino, Domino Foods Inc., Iselin, NJ) and imitation vanilla extract were purchased at a local grocery. Sucralose was provided by Bulk Supplements (Henderson, NV). Stevia was provided by Tate & Lyle (Tasteva, Decatur, IL) and contained 95% steviol glycosides according to the manufacturer. Monk fruit extract was provided by Tate & Lyle (PureFruit Select) and contained at least 48% mogroside V on a weight basis according to the manufacturer.

Protein content was maintained at 11% wt/wt protein for all model foods and each was formulated to standardize the perception of vanilla flavor (Campbell et al., 2016). Stock solutions of WPI were prepared by dispersing WPI powder in deionized water via stirring at room temperature (herein 22 ± 2°C) for 4 hr to achieve complete hydration. During formulation, pH was adjusted to 6.9 using food grade 1 N NaOH or HCl (Sigma-Aldrich, St. Louis, MO). The solutions were brought to final mass with deionized water and held at 4°C overnight. Prior to heating, solutions were equilibrated to room temperature for 1 hr.

TABLE 1 Sweetener concentration (% wt/wt) ranges used for each texture in magnitude estimation scaling

	Thin fluid (F1)	Thick fluid (F2)	Semisolid (SS)
Sucrose	2.0–12.6	2.0–12.6	2.0–12.6
Sucralose	0.007–0.059	0.009–0.076	0.009–0.076
Stevia	0.030–0.158	0.030–0.158	0.030–0.158
Monk fruit	0.029–0.427	0.029–0.427	0.029–0.427

Solution aliquots of 30 g were poured into glass tubes (19 mm internal diameter × 180 mm length) coated with food grade silicone mold release (Price Driscoll Corp., Waterford, CT) and closed with rubber stoppers on the bottom. Model food texture was modulated by varying heating time (0–15 min at 80°C) to generate thin fluid (F1), thick fluid (F2), and semisolid (SS) structures. Due to the viscosity of sucrose as a bulk ingredient, heating time was adjusted to compensate for the higher viscosity of sucrose samples. Tubes were covered with aluminum foil to limit evaporation during heating. After heating, samples were cooled at room temperature for 1 hr and held overnight at 4°C. Final samples exhibited no significant differences in apparent viscosity (fluids) or complex viscosity for the semisolids (data not shown) among all sweetener treatments. Each treatment was equilibrated to room temperature prior to sensory evaluation.

2.2 | Sweetness power functions

All sensory testing was performed at North Carolina State University in compliance with the North Carolina State University Institutional Review Board for Human Subjects. Power functions for sweeteners in all three texture types were generated using MES (Li et al., 2015; Meilgaard et al., 2007). Panelists ($n = 12$, 7 females, 5 males, ages 22 to 41 years) participated in 2.5 hr of MES training sessions and were confirmed to demonstrate replicable ratio scaling with a range of sweetened solutions and whey protein samples prior to data collection. The MES scaling method was used to create power function curves using 6% wt/wt sucrose as a reference intensity 100, and scale anchors of 3 and 9% wt/wt sucrose representing intensities of 50 and 150, respectively. Panelists were instructed to refer to these references as they tasted each test treatment.

Seven sweetener concentrations of each treatment were formulated using geometric scaling factors across the concentration ranges listed in Table 1. Each range was selected to represent the lowest detectable sweetness and to cover the sweetness intensity range of the references. No more than 12 samples were presented during any session. Panelists were presented with samples coded with 3-digit numbers and presented in a random order, and asked to scale maximum perceived sweetness to the reference solutions. Rests of 4 min were enforced between each sample and panelists were instructed to rinse their mouth with water and take a bite of an unsalted cracker between samples. Each treatment was evaluated in triplicate and data were collected using Compusense Cloud (Compusense, Guelph, Canada).

2.3 | Determination and confirmation of iso-sweetness

Iso-sweetness across textures determined using MES was confirmed using a directional paired comparison test (2-Alternative Forced Choice, 2-AFC) (ASTM, 1991). Samples were presented in lidded 60 mL soufflé cups with 3-digit codes using a balanced presentation order. Panelists ($n = 30$) were instructed to taste each set of two samples in the presented order and to select the sample they believed was sweeter. A 5 min rest period was enforced between each set of samples, during which panelists were instructed to rinse their mouth with water and take a bite of an unsalted cracker. Data for 2-AFC testing were collected on iPads using Compusense Cloud (Compusense, Guelph, Canada).

2.4 | Temporal profiling via temporal dominance of sensations

Perception of dominant attributes over time was determined using TDS with a trained panel ($n = 9$; 6 females, 3 males, ages 23–54 years) (Pineau et al., 2009). Confirmed iso-sweet sweetener concentrations were used to ensure that the sweet taste intensity was consistent among different textures. Panelists were provided 20 mL samples presented in lidded 59 mL soufflé cups with 3-digit codes using a balanced presentation order. Panelists were instructed to put the entire sample in their mouth, start the timer, and to select the most dominant attribute. A combination of taste, flavor, and oral-tactile sensational attributes were presented to the panelists, including sweet taste, bitter taste, metallic, sweet aromatic flavor, cardboard flavor, and astringency. Panelists could also select "none" as an option. Panelists were prompted to expectorate the sample after 15 s and to evaluate the sample over the course of 192 s. Each sample was evaluated by each panelist in triplicate. Data were collected on iPads using Compusense Cloud (Compusense, Guelph, Canada).

2.5 | Statistical analysis

Hypothetical iso-sweet concentrations were generated for each treatment and sweetener combination by graphing the dose-response power functions from MES and calculating the sweetener concentration required for intensity equivalent to the reference 100 (i.e., 6% wt/wt sucrose). Sweetener intensity ranges relative to sucrose were calculated as the ratio of sucrose to sweetener at iso-sweet concentrations. Statistical analysis of power function slopes and equivalent sweetener concentrations was performed using two-way ANOVA accounting for texture × sweetener interactions (JMP v12.0, SAS, Cary, NC) followed by a post hoc means separation using Tukey's HSD ($\alpha = 0.05$). Statistical significance of 2-AFC paired comparison tests was determined based on the minimum number of correct judgments for significance at $\alpha = 0.05$ (Meilgaard et al., 2007). Temporal data from TDS were presented as curves where attribute dominance rates were plotted over time using XLSTAT v18.01 (Addinsoft, New York, NY). A smoothing function within the software was used to construct the curves. Significance was calculated at 95% confidence with a chance rate of 14.3% based on seven attributes.

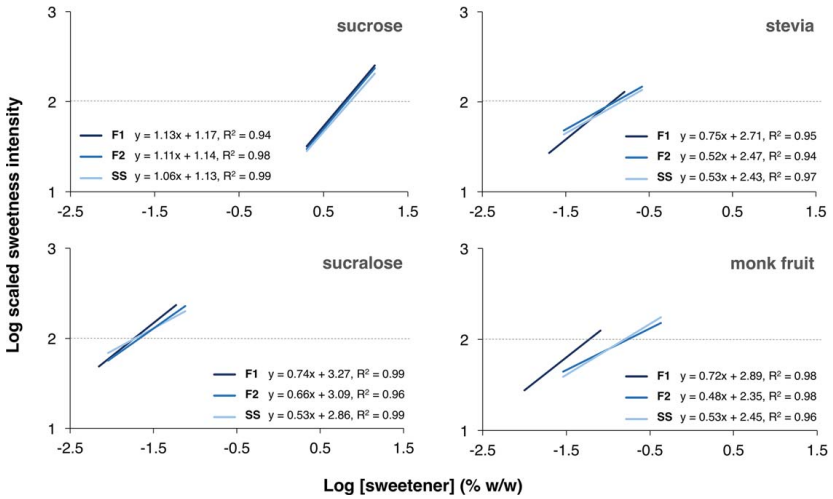

FIGURE 1 Sweetness power functions of sweeteners in thin fluid, thick fluid, and semisolid textures. Power functions are represented as the best fit line of three independent replications across seven sweetener concentrations for thin fluid (F1), thick fluid (F2), and semisolid (SS) textures. The dotted line indicates the scaled sweetness intensity of 6% wt/wt sucrose used as the reference 100

3 | RESULTS

3.1 | Sweetness power functions

MES is a ratio scaling technique frequently employed to generate power functions (i.e., dose-response functions) of sweeteners. This method is advantageous in that it yields a scale with true ratio properties, no arbitrarily limited endpoints, and scalable data that can be compared across studies (Cardello et al., 1999). Three types of sweeteners were used in this study: a caloric sweetener (sucrose), artificial non-nutritive sweetener (sucralose), and two natural non-nutritive sweeteners (stevia and monk fruit extracts). Power functions for each texture and sweetener type are shown in Figure 1. As expected, an intensity difference of several orders of magnitude was observed between sucrose and the non-nutritive sweeteners. Sweetener intensity ranges relative to sucrose were calculated as the ratio of sucrose to sweetener at iso-sweet concentrations. Relative to sucrose, sweetness intensity across all textures was 45 to 54 times greater for stevia, 280 to 348 times greater for sucralose, and 35 to 100 times greater for monk fruit. Additionally, there was a significant texture and sweetener interaction on power functions for all sweeteners with the exception of sucrose ($p < .05$).

Power functions display a linear relationship between magnitude of physical stimulus and a perceived magnitude, and the exponent or slope of the power function denotes the rate of change in perceived sweetness as a function of concentration (Cardello et al., 1999; Lawless & Heymann, 2010). A significant sweetener and texture interaction on slope of the power function was observed for all artificial sweeteners (Table 2). Sucralose exhibited the most marked texture and sweetener interaction, trending toward a decreased slope with increasing thickness from thin fluid (F1 = 0.74) to thick fluid (F2 = 0.66) and semisolid samples (SS = 0.53). This indicates more sweetener is required for equivalent sweet taste as texture is altered. Additionally, all non-nutritive sweeteners had significantly lower slopes than sucrose at all textures, which has previously been reported for non-nutritive sweeteners in skim chocolate milk and yogurt (Li et al., 2015; Reis et al., 2011). The smaller exponent suggests that even more sweetener would be needed for equal perception of sweetness at sucrose concentrations greater than 6% wt/wt. Stevia and monk fruit had similar power functions, which has previously been reported for these sweeteners in skim chocolate milk (Li et al., 2015). For samples sweetened with stevia and monk fruit extracts, a significant texture and sweetener interactions on the power function slope were only observed between thin fluid and thick fluid/semisolid.

The dotted line in Figure 1 indicates a scaled sweetness intensity equivalent to a 6% wt/wt sucrose solution. The intersection of individual power functions with this line represents the sweetener concentration required for iso-sweetness with a 6% sucrose reference solution; these values are shown in Table 3. Although the slope of the power function

TABLE 2 Power function slopes of sweeteners in thin fluid, thick fluid, and semisolid textures

	Thin fluid (F1)	Thick fluid (F2)	Semisolid (SS)
Sucrose	1.13 ± 0.05 a	1.06 ± 0.09 a	1.07 ± 0.11 a
Sucralose	0.737 ± 0.005 b	0.655 ± 0.040 c	0.527 ± 0.024 cd
Stevia	0.752 ± 0.062 b	0.519 ± 0.089 cd	0.526 ± 0.047 cd
Monk fruit	0.720 ± 0.012 c	0.479 ± 0.034 d	0.532 ± 0.025 cd

Numbers are presented as arithmetic means ± standard deviation. Letters indicate significant differences among means as determined by two-way ANOVA followed by Tukey's HSD post hoc comparisons ($\alpha = .05$).

TABLE 3 ISO-sweet concentrations (% wt/wt) of sweeteners in thin fluid, thick fluid, and semisolid textures

	Thin fluid (F1)	Thick fluid (F2)	Semisolid (SS)
Sucrose	5.95 ± 0.13 b	6.02 ± 0.46 b	6.54 ± 0.23 a
Sucralose	0.0186 ± 0.0019 g	0.0215 ± 0.0090 g	0.0188 ± 0.0035 g
Stevia	0.123 ± 0.004 de	0.112 ± 0.009 e	0.146 ± 0.010 cd
Monk fruit	0.059 ± 0.003 f	0.171 ± 0.065 c	0.157 ± 0.004 c

Iso-sweet equivalencies based on 6% wt/wt sucrose in water as a reference. Numbers are presented as arithmetic means ± standard deviation. Letters indicate significant differences among means as determined by two-way ANOVA followed by Tukey's HSD post hoc comparisons ($\alpha = .05$).

did not change among textures for sucrose, the texture and sweetener interaction had a significant influence on iso-sweet concentration. More sucrose was required for iso-sweetness only when transitioning between fluids (F1 = 6.0; F2 = 6.0) and the semisolid (SS = 6.5). Despite the significant reduction in slope at the fluid-semisolid transition for sucralose samples, iso-sweet concentrations were not statistically distinct. This is not surprising as the power functions for the three textures intersect near the 6% sucrose equivalency line. Per these power functions, this suggests that texture would have a stronger effect at sucrose equivalencies other than 6% wt/wt. Stevia samples trended toward the same fluid-semisolid transition although only the thick fluid (F2 = 0.11) and semisolid textures (SS = 0.15) were statistically distinct. Monk fruit samples exhibited a marked increase in iso-sweet concentration from the thin fluid (F1 = 0.06) to thick fluid and semisolid (F2 = 0.17; SS = 0.16). As an extra substantiation step prior to TDS analysis, iso-sweet concentrations from MES were confirmed using directional 2-AFC paired comparison tests with naïve panelists ($n = 30$). The results (data not shown) confirmed no significant differences among paired comparisons across textures for each sweetener, corroborating that the sweetness equivalencies determined from MES power functions were iso-sweet across textures.

3.2 | Temporal dominance of sensations

Due to the different temporal profile of sweetness for these sweeteners, TDS was used to evaluate the progression of perceived dominant attributes over time. With TDS, panelists are presented with a selection of attributes and asked to select the most dominant or striking attribute at any given time (Pineau et al., 2009). The resulting data demonstrate the evolution of several attributes to show product perception pattern over time, providing qualitative insight into sensations that consumers perceive over time (Le Révérend et al., 2008). In studying texture-taste interactions, TDS is useful because taste, texture, and flavor attributes can be presented simultaneously, providing insight into what informs consumer responses.

Considering all treatments, the samples were characterized by sweet and bitter tastes, astringency, metallic flavor, sweet aromatic flavor, and cardboard flavor. Sucrose samples were characterized by dominant sweetness progressing to cardboard flavor and astringency after expectoration (Figure 2). Thin fluid and semisolid samples also showed dominant sweet aromatic sensations at the same time as sweetness. Increasing thickness from thin fluid to thick fluid and semisolid structures extended the duration that astringency was perceived as a dominant attribute and delayed the onset of "no dominant sensation."

Of the three non-nutritive sweeteners in this study, sucralose temporality was most similar to sucrose. Sucralose samples were characterized by sweetness progressing to metallic, then astringency (Figure 3). Cardboard flavor was dominant for a short amount of time in the fluid, which was also observed with sucrose samples. The onset time of astringency as a dominant attribute did not appear to differ among textures, but the total duration increased with increasing texture. Bitterness was not perceived as a dominant attribute for any of the sucrose or sucralose-sweetened textures.

Samples sweetened with stevia were characterized by early sweet taste and bitterness followed by metallic taste and astringency (Figure 4). A higher percentage of panelists perceived bitterness as dominant compared to sweetness for the thin fluid, whereas the reverse was true for the thick fluid and semisolid. The onset of metallic taste as a dominant attribute was the same for all three textures, but the total duration metallic taste was perceived as dominant decreased with increasing texture. The temporal profiles of samples sweetened

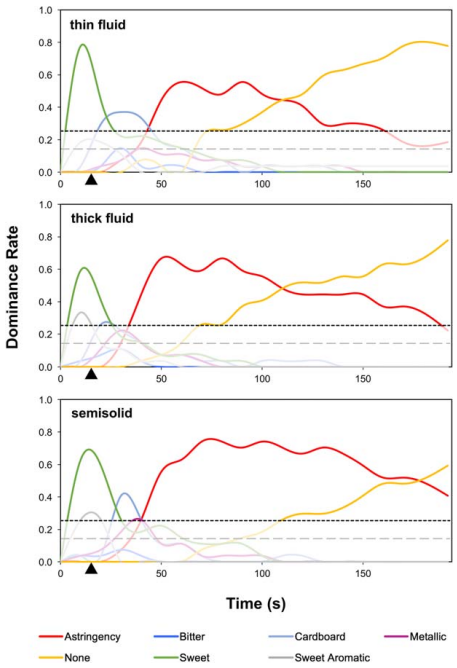

FIGURE 2 Temporal attribute dominance of thin fluid, thick fluid, and semisolid textures sweetened with sucrose. Dotted line indicates significance level ($\alpha = 0.05$) and dashed line indicates chance level. The triangle on the *x*-axis indicates time when samples were expectorated. Data are faded below the significance line to facilitate comparisons among treatments

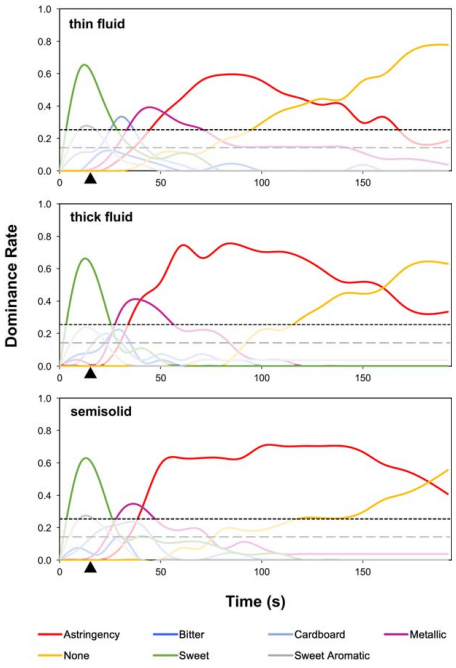

FIGURE 3 Temporal attribute dominance of thin fluid, thick fluid, and semisolid textures sweetened with sucralose. Dotted line indicates significance level ($\alpha = 0.05$) and dashed line indicates chance level. The triangle on the *x*-axis indicates time when samples were expectorated. Data are faded below the significance line to facilitate comparisons among treatments

with monk fruit extract were similar to stevia, with a progression from sweet to bitter to metallic to astringency for all textures (Figure 5). A texture effect was less pronounced for samples sweetened with monk fruit, although the sweetness dominance rate appeared to increase with increasing thickness and the duration of metallic taste dominance was longer in the semisolid samples compared to the fluids.

4 | DISCUSSION

Over consumption of sucrose is a global problem that drives sugar reduction in food product development. This provides an opportunity for reformulation with alternative sweeteners to reduce the caloric density of foods; however, they can have significant effects upon sweetness perception and final food texture. Texture alone has health implications. Increased viscosity and thicker textures have been implicated in satiation and satiety, where thicker beverages or transitions from liquid to semisolid attenuate subjective ratings of hunger and reduce ad libitum intake (de Wijk et al., 2008; Mars et al., 2009).

Previous reports indicate suppressed flavor and taste perception with increasing viscosity of fluids (Arabie & Moskowitz, 1971; Hollowood et al., 2002). However, the influence of texture across fluids and semisolids has not been explored. The traditional approach of modifying texture with added ingredients is likely to introduce other confounding variables. Therefore, we developed the model system in the present study based on a single set of molecules, where the generation of distinct textures was achieved by adjusting total heating time. Heating the samples leads to the formation of primary protein aggregates and changing heating time alters the degree of protein aggregation, and thus viscosity and elasticity (Ikeda, 2003). Sensory texture attributes of the three textures used in this study have been previously characterized and were significantly differentiated in terms of in mouth viscosity ($F1 = 1.2$, $F2 = 3.5$, and $SS = 6.6$) using descriptive analysis (Campbell et al., 2016). For simplicity of discussion in the present study, sample viscosities can be thought of in terms of fluid milk (fluid), drinkable yogurt beverage (thick fluid), and spoonable pudding (semisolid). Additionally, all sweeteners used in the study were selected on the basis of high heat stability to mitigate thermal degradation as a confounding variable.

Based on the literature, increasing sample viscosity or forming a semisolid in the current study was expected to decrease sweetness perception and increase sweetener concentration necessary to achieve

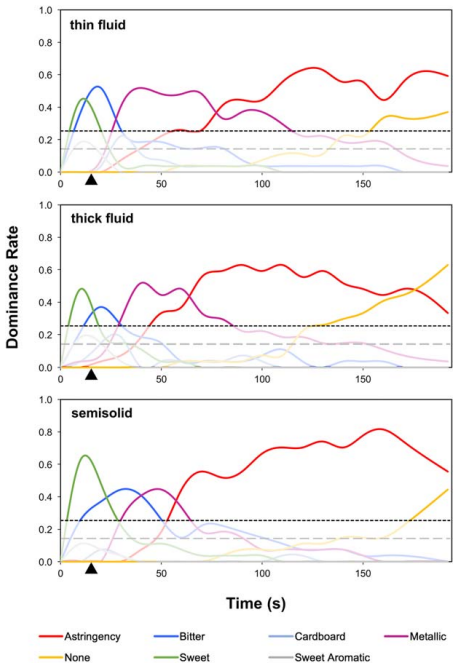

FIGURE 4 Temporal attribute dominance of thin fluid, thick fluid, and semisolid textures sweetened with stevia extract. Dotted line indicates significance level ($\alpha = 0.05$) and dashed line indicates chance level. The triangle on the *x*-axis indicates time when samples were expectorated. Data are faded below the significance line to facilitate comparisons among treatments

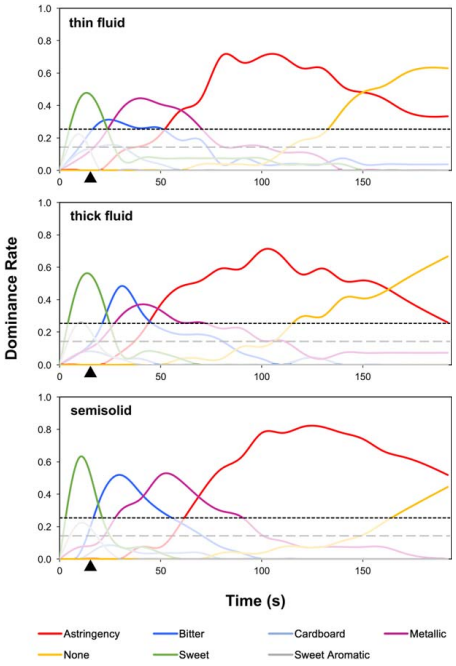

FIGURE 5 Temporal attribute dominance of thin fluid, thick fluid, and semisolid textures sweetened with monk fruit extract. Dotted line indicates significance level ($\alpha = 0.05$) and dashed line indicates chance level. The triangle on the *x*-axis indicates time when samples were expectorated. Data are faded below the significance line to facilitate comparisons among treatments

iso-sweetness. All samples exhibited a significant texture sweetener interaction on either the iso-sweet concentration or slope of the power function. A statistically significant effect on iso-sweet concentration was observed for all sweeteners except for sucralose. From a practical standpoint, this corresponded to requiring 10% more sucrose, 19% more stevia, and 166% more monk fruit when comparing the thin fluid to the semisolid textures. In the case of sucrose and stevia, differences in iso-sweet concentrations were observed only between the thin fluid/thick fluid and semisolid, whereas for monk fruit it was between thin fluid and thick fluid/semisolid. Also, even though no effect of texture was observed on iso-sweetness of sucralose, the slope of the power function did decrease with increasing thickness. This would suggest that at higher sweetness equivalencies (i.e., greater than 6% wt/wt sucrose), the effect of texture would be more pronounced and more sucralose would be needed for equivalent sweetness. A similar reduction in sweetness power function slope was observed when increasing the complexity of a system from water to skim milk to skim chocolate milk (Li et al., 2015).

Sucralose has been previously reported to exhibit a temporal profile more similar to sucrose than other non-nutritive sweeteners (Morais et al., 2014, Zorn et al., 2014). The main difference was the appearance of metallic taste as a significant attribute after sweet taste for sucralose, which was not observed for sucrose. Dominant bitterness was not observed for sucralose, unlike stevia and monk fruit. These natural non-nutritive sweeteners had similar temporal profiles, and texture altered the power functions and iso-sweet concentrations in similar ways. Dominant bitter and metallic tastes have also been reported for these sweeteners in water, fluid fruit juice, and skim chocolate milk (Cardoso & Bolini, 2008; Li et al., 2015; Reyes et al., 2017).

Studies by Paixão et al. (2014) and Li et al. (2015) reported that the addition of fat or dairy and cocoa solids alters the dose-response function. The results of this study build on these by showing that texture *alone* can influence sweetness without modifying formulation. The observed decrease in sweetness perception with increasing viscous and elastic elements of texture could potentially be explained with a diffusion model strictly on the basis of rheological properties. Tastant molecules are solvated in saliva and travel through a diffusion-controlled regime at the surface of the tongue to reach taste receptors; the overall flux (i.e., amount of tastant reaching target per area per time) is one parameter that governs perceived intensity (Aubert et al., 2016; Kokini, 1987; Pfeiffer et al., 2000). As sample viscosity is increased, the diffusion rate of molecules is reduced, thus lowering flux rate and perceived intensity.

Alternatively, the type of underlying structure—i.e., to say *how* viscosity is increased—may also be an important consideration. In the case of polymer-thickened fluids forming random coil networks, perception of sweetness is predicated on polymer concentration relative to a critical overlap concentration, c^* (Baines & Morris, 1987; Cook et al., 2002; Hollowood et al., 2002). At concentrations above c^*, polymers exhibit overlapping hydrodynamic volumes; this corresponds to a large increase in solution viscosity. It is thought that above c^*, inefficient mixing of food and saliva suppresses flux of molecules to receptors (Baines & Morris, 1987). In the present study, the fluid networks are thickened by protein aggregates, with a higher degree of aggregation for the thick fluid. It is possible that a reduction in sweetness or power function slope for the thick fluid corresponds to overlapping hydrodynamic volumes (i.e., exceeding an effective c^*). Additionally, the semisolid texture is comprised of an entangled protein network, which could entrap sweetener molecules within the matrix and further slow the release of molecules to receptor.

Although taste suppression above c^* would be detrimental in terms of reducing sucrose, it is unknown if this reported reduction is simply a reduction in maximum intensity or a prolonging of the temporal profile. As structure is changed from a fluid to a semisolid, there is a corresponding increase in sensory adhesiveness (Campbell et al., 2016), suggesting a possible role of mucoadhesion. The concept of inefficient mixing coupled with mucoadhesion would suggest more tastant molecules are around to bind receptors, which would be expected to alter the temporal profile. However, TDS only provides qualitative data about the product as a whole; single attribute time intensity testing would be needed to determine if attribute intensity persists.

The reduction in iso-sweet concentration and power function slope with increasing texture could be associate with prolonged sensation of astringency in these samples. Astringency in whey protein fluids

is typically associated with low pH but can also be a problem at high protein concentrations and neutral pH due to mucoadhesion (Beecher et al., 2008; Withers et al., 2013). Moreover, longer heating of whey proteins is reported to increase astringency and mouthcoating (Bull et al., 2017). At pH 6.9 and 11% wt/wt protein, astringency of the samples in this study could be explained by these mechanisms. Reports that low shear viscosity controls taste suppression also give credence to the mucoadhesion mechanism (Aubert et al., 2016).

It is important to note that with TDS, panelists select only one dominant attribute at a time. As a result, dominance of one attribute does not indicate the absence of another. This may explain the decrease in duration of metallic taste dominance with increasing thickness for stevia samples. Metallic taste could still be present, but astringency is perceived as more dominant. Another important consideration not directly addressed in this study is the role of oral processing in perception. The way in which a material is transported around the oral cavity influences the perception of flavor and texture attributes (de Wijk et al., 2003). As a result, the oral processing activities, movements, and manipulations used to process fluid and semisolids may differ, changing the rate at which a sweetener reaches a detector.

5 | CONCLUSIONS

This study established the perceived sweetness intensity of four common sweeteners in different food textures. A significant texture-sweetener interaction on sweetener concentration required for sweet taste equivalency was observed. Texture did not alter the power function slope for sucrose-sweetened samples, but a reduced power function slope with increasing thickness was observed for all non-nutritive sweeteners. All non-nutritive sweeteners had similar power function slopes at most textures, and all were significantly lower than sucrose. However, the iso-sweet concentrations were different, with different effects of texture across all sweeteners. This indicates that sugar reduction strategies are not "one size fits all" in terms of product texture and desired sweetness level. These differences are likely related to the effect of texture on perceived dominant attributes, where some attributes tended to be perceived as dominant for a longer amount of time with increasing thickness, and others peaked at different times. Differences in perceived sweetness were related to each texture having distinct temporal profiles. Most notably, increased thickness was associated with prolonged sensation of astringency. The results of this study indicate that food matrix, sweetener type, and sweetener usage level need to be considered when attempting to modify texture or reduce sucrose. Further research should explore the relatively unknown role of mucoadhesion in taste perception or provide mechanistic insight into how texture modifies taste and flavor perception.

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CHAPTER 5. ROLE OF MUCOADHESIVE HYDROCOLLOIDS IN TASTE AND TEXTURE PERCEPTION

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1. ABSTRACT

Mucoadhesion of food components to oral mucosa during consumption is reported to play a role in taste and texture perception. Mucoadhesive strength depends on viscosity and molecular interactions with molecules along the oral mucosa, namely mucins. In this study, poor (starch) and strong mucoadhesives (λ–carrageenan) were used to determine how mucoadhesion, viscosity level (4–6 mPa·s, 25–30 mPa·s, and 50–60 mPa·s at 50 1/s), and complexity of system (aqueous, skim milk, or whole milk) influence sensory taste and texture. Rheological flow profiles showed shear thinning in all samples, but compared to starch, carrageenan had much higher low shear viscosity and lower high shear viscosity in skim milk and whole milk despite being matched at the oft-reported shear rate of the mouth. Sensory analysis revealed a significant effect of viscosity level but a weak effect of hydrocolloid type. Normalized against perceived sensory consistency, no effect of hydrocolloid type was observed. Additionally, increasing viscosity several orders of magnitude did not suppress perceived sweet taste. Temporal dominance of sensations shows that samples exhibit similar sensory profiles, although the addition of hydrocolloids enhanced dominance of creaminess even in samples without fat. Hydrocolloid type did not significantly influence mouthcoating or the persistence of astringency. The results suggest that in fluid systems with viscosity levels typically found in beverages, type of hydrocolloid did not have a significant effect on taste perception.

2. INTRODUCTION

Hydrocolloids are commonly added to fluid products to increase viscosity, alter mouthfeel, enhance stability, or to replace attributes that are lost as part of decreasing caloric density of foods, such as sugar or fat reduction. Increased viscosity can be desirable to enhance stability or alter texture perception, but viscosity has long been reported to suppress taste and flavor (Baines & Morris, 1987; Christensen, 1980; Moskowitz & Arabie, 1970). The degree of viscosity increase required for taste suppression in unclear, with Pangborn et al. (1973) reporting suppression at 16 mPa·s and Ferry et al. (2006) reporting suppression at nearly 280 mPa·s. Additionally, early investigations into texture-taste interactions suggested viscosity influences taste and implied that rules may not be general and may be polysaccharide type-dependent (Christensen, 1980; Pangborn, Trabue, & Szczesniak, 1973).

Despite awareness of viscosity-taste interactions for decades, the mechanisms are still unknown. One hypothesis is that viscosity suppresses taste by restricting diffusion of molecules to receptors. Alternatively, taste suppression has also been explained on the basis of microstructure at critical overlap, c* (Baines & Morris, 1987; Cook, Hollowood, Linforth, & Taylor, 2002, 2003). The c* occurs describes the concentration at which a random coil hydrocolloid solution transitions from dilute to overlapping hydrodynamic radii and is typically associated with a marked increase in solution viscosity. Numerous studies have shown that at concentrations exceeding c* (which depends on the properties of the hydrocolloid in question) taste is suppressed (Baines & Morris, 1987; Cook et al., 2002; Han et al., 2014). Considering c* applies to polymeric systems, it is unknown if molecular structuring could alter taste or texture in more structurally complex systems containing molecules across multiple length scales.

In order to explain how food structure modulates taste, numerous models have been proposed to describe and predict mass transfer of sapid molecules during oral processing (Aubert, Lima, & Le Révérend, 2016; Le Révérend, Norton, & Bakalis, 2013; Pfeiffer, Boulton, & Noble, 2000). The mouth can be modeled as a well-mixed bulk region comprising most of the volume of the oral cavity and therefore tastant mass transfer occurs rapidly with fluid foods (Aubert et al., 2016; Le Révérend et al., 2013). A thin salivary pellicle with thickness 70–100 μm sits on top of oral surfaces (Collins & Dawes, 1987; Gibbins & Carpenter, 2013). Due to the non-smoothness of oral surfaces, fluid flow is restricted and therefore encounters low shear rates (Aubert et al., 2016). This may explain strong correlations between low shear viscosity and taste perception (He, Hort, & Wolf, 2016). The interfacial region between the bulk and pellicle is governed by Fickian mass transfer based on a concentration gradient. Moreover, the thickness of this salivary layer has been shown to be dependent on tastant delivery velocity in rats, where higher velocities reduced the thickness of this layer and should therefore enhance mass transfer (DeSimone, 2017; Gibbins & Carpenter, 2013). Therefore, viscosity and tastant mass transfer are important for taste perception. Viscosity in this sense is not a singular property, as most hydrocolloids exhibit shear-thinning behavior and therefore viscosity is dependent on shear rate. A shear rate of 50 1/s is often reported as the shear rate in the mouth due to correlations between viscosity and sensory thickness, but the biophysics of oral cavity suggest that fluids

experience a range of shear rates during oral processing (Kokini, 1987; Shama & Sherman, 1973; Stokes, Boehm, & Baier, 2013; Wood, 1968). The viscosity of non-Newtonian fluids depends on shear rate and a number of studies have focused on understanding which portion of the flow profile is responsible for perception (Aubert et al., 2016; He et al., 2016; Stokes, Macakova, Chojnicka-Paszun, de Kruif, & de Jongh, 2011). The summation of these studies indicates that the entire shear rate range provides a more comprehensive understanding of viscosity-taste-texture interactions.

More recently the concept of mucoadhesion – the capacity of a hydrocolloid to bind mucosal tissues in the body – has been implicated in modulating taste perception (Cook, Bull, Methven, Parker, & Khutoryanskiy, 2017; Cook, Methven, Parker, & Khutoryanskiy, 2018; Cook, Woods, Methven, Parker, & Khutoryanskiy, 2018). The overall binding strength between mucoadhesive and oral mucosa is due in part to viscosity (enhancing ability to remain in contact with mucosa), but also due to molecular interactions between mucoadhesive and mucosal tissues. Mucoadhesion has long been leveraged by the pharmaceutical industry to modify molecular properties to control delivery of active ingredients to cells (Smart, 2005). Similar concepts may apply to the perceived taste and flavor of food products during consumption (Cook et al., 2017).

Mucoadhesion has rarely been addressed directly in food science literature, but several recent studies have touched on mucoadhesion in regard to taste and texture perception using a variety of *in vitro* and *in vivo* approaches. Withers et al. evaluated how the two principle milk proteins (casein and β–lactoglobulin) bind to porcine mucosa, reporting that although both molecules exhibited mucoadhesive properties, caseins bind more strongly than β–lactoglobulin due to increased solution viscosity and lower overall charge (Withers, Cook, Methven, Gosney, & Khutoryanskiy, 2013). However, no sensory analysis was provided to determine if disparate *in vitro* mucoadhesion translates to detectable differences in humans. Cook et al. reported that carboxymethyl cellulose (CMC) exhibited greater *in vitro* mucoadhesion than starch, which correlated with increased sensory mouthcoating and decreased perceived temporal salty taste intensity (Cook, Woods, et al., 2018). It should be noted that salt concentration was adjusted to compensate for the sodium ions present in CMC; therefore, it is difficult to determine if the taste effect was simply due to the lower sodium concentration. An effect of hydrocolloid type was also reported in a study exploring

mucoadhesive interactions between mucins and pectin, CMC, or sodium alginates (Ali & Bakalis, 2011). A significant effect of solution viscosity on binding affinity was recorded, similar to the results reported by Withers et al. (Ali & Bakalis, 2011; Withers et al., 2013).

In addition to influencing taste and texture perception, mucoadhesion is also reported to play a role in the oral-tactile perception of astringency or mouth-drying. Whey protein solutions heated for longer times (and thus a greater extent of protein aggregation and a concomitant increase in mean particle size) were reported by a sensory panel as being more astringent and mouthcoating (Bull et al., 2017). Ferry et al. reported that mouthfeel, flavor, and taste in a model savory sauce were dependent on the type of thickener due to the underlying microstructure (Ferry et al., 2006). Granular starch systems mixed more efficiently with artificial saliva than polymeric HPMC, which the authors used as the explanation for greater taste suppression in HPMC at high viscosity.

These studies indicate that mucoadhesion and microstructure play important roles in the perception of taste, texture, and oral-tactile sensations. However, there is still a gap related to oral perception and how different mucoadhesive ingredients influence the perception of taste and mouthfeel. Considering mucoadhesion is a balance between the macroscopic (i.e., viscosity) and microscopic (i.e., molecular interactions between mucins and mucoadhesives) scales, it is unknown if a certain viscosity level needs to be reached in order to alter taste, or if compositional differences among food products influence the effect. Therefore, we hypothesize that transitions in structural properties due to different hydrocolloids, different viscosity levels, and interactions with oral surfaces will alter cognition of taste. The objectives of this study were to compare the effects of low and high mucoadhesive hydrocolloids (carrageenan and starch) on perceived taste and texture at different viscosity levels (4–6 mPa·s, 25–30 mPa·s, and 50–60 mPa·s at 50 1/s) in three separate fluid systems (water, skim milk, and whole milk). An additional objective was to evaluate how these systems alter temporal perception during consumption.

3. MATERIALS & METHODS

3.1. Materials

Skim milk powder (SMP) and whole milk powder (WMP) were provided by Fonterra (Auckland, New Zealand). Food grade λ–carrageenan reported as medium viscosity was

provided by Danisco, (Dupont, Copenhagen, Denmark) and enzymatically modified tapioca starch was provided by Ingredion (Westchester, IL, USA). Food grade NaOH and HCl were purchased from Sigma-Aldrich and sucrose from a local grocery.

3.2. Sample Preparation

Stock solutions of SMP and WMP were formulated at matched final protein of 3.0% w/w, which represented 12.25% total solids for WMP and 9.21% total solids for SMP. Stock solutions were prepared by dispersing respective powders into deionized water at 600 rpm for 60 s, then in a 55 ˚C water bath for 120 min. Solutions were stirred using an overhead mixer (Heidolph RZR, Schwabach, Germany) with attached blade impeller (25 mm diam.) at 400 rpm to ensure complete hydration. Separately, starch and carrageenan stock solutions were prepared by dispersing powder into deionized water at 600 rpm in an 85 ˚C water bath for 30 min using the aforementioned overhead mixer with attached blade impeller. Samples were then mixed with the necessary amount of WMP or SMP stock solutions and deionized water to reach desired final total solids. When necessary, pH was adjusted to 6.9 ± 0.1 with 1 N HCl or NaOH. All samples were cooled in an ice bath and stored overnight at 4 ˚C before use. Samples for sensory analysis were prepared fresh daily and used within 24 hours, all other samples included 200 ppm sodium azide as a microbial inhibitor and were used within 4 days.

3.3. Rheology

Rheological flow profiles were determined using a controlled stress rheometer (Anton Paar Physica MCR 301; Graz, Austria). A combination of geometries and methods were used to evaluate both low shear and high shear regions of sample flow profiles. Low shear measurements were performed using couette geometry (27 mm diam.) at 20 ± 0.1 ˚C. Samples were pre-sheared at 100 1/s for 60 s and quiescently held for another 60 s immediately prior to measurements. Logarithmic shear rate sweeps were performed from 0.01 to 300 1/s. Data with torque values at or below the manufacturer reported sensitivity were omitted.

Table 5.1. Formulations for hydrocolloid model systems in water, skim milk powder, and whole milk powder at three viscosity targets.

Continuous phase	Thickener	Viscosity	WMP (% TS)	SMP (% TS)	Sucrose (g/kg)	Starch (g/kg)	CG (g/kg)
Water	Starch	Low	0	0	30.0	14.0	0
Water	Starch	Medium	0	0	30.0	32.5	0
Water	Starch	High	0	0	30.0	44.5	0
Water	CG	Low	0	0	30.0	0	0.86
Water	CG	Medium	0	0	30.0	0	4.80
Water	CG	High	0	0	30.0	0	7.80
SMP	Starch	Low	0	9.21	30.0	8.0	0
SMP	Starch	Medium	0	9.21	30.0	22.0	0
SMP	Starch	High	0	9.21	30.0	31.0	0
SMP	CG	Low	0	9.21	30.0	0	0.20
SMP	CG	Medium	0	9.21	30.0	0	0.48
SMP	CG	High	0	9.21	30.0	0	0.80
WMP	Starch	Low	12.25	0	30.0	6.40	0
WMP	Starch	Medium	12.25	0	30.0	17.5	0
WMP	Starch	High	12.25	0	30.0	25.5	0
WMP	CG	Low	12.25	0	30.0	0	0.11
WMP	CG	Medium	12.25	0	30.0	0	0.32
WMP	CG	High	12.25	0	30.0	0	0.55

Viscosity targets represent 4–6 mPa·s (low), 25–30 mPa·s (medium), and 50–60 mPa·s (high) at 50 1/s. CG: λ–carrageenan; WMP: whole milk powder; SMP: skim milk powder; TS: total solids.

Narrow gap rheology was performed using parallel plate geometry (20 mm diam.) at a 100 μm gap per the method described by Davies & Stokes (2008). Viscosity of a 1000 cP oil standard at gap distances of 800, 600, 400, 200, 100, and 50 μm was measured and non-linear regression was used to determine the gap error (Kravchuk & Stokes, 2013). Logarithmic shear rate sweeps were performed from 0.1 to 30,000 1/s at 20 ± 0.1 ˚C with a 5 cP oil standard applied at the sample-air interface. Measured viscosity was corrected for gap error using Eq. 1.

$$\eta_{corrected} = \eta_{measured} * \left(1 + \frac{gap\ error}{gap\ distance}\right) \quad (1)$$

3.4. Descriptive sensory analysis

Descriptive sensory analysis of taste, flavor, and texture attributes was performed using a trained panel (n = 14) of members of an existing descriptive analysis panel at the Fonterra Research and Development Centre (Palmerston North, New Zealand). Data were collected over three separate sessions, with panelists evaluating samples in separate booths under white light. Each panelist had greater than 50 hours experience on descriptive analysis of taste and texture using the quantitative descriptive analyses (QDA) method. Attributes shown in Table 5.2 were based on established lexica for fluids. All attributes were reviewed for appropriateness by the panel during a training session using samples representative of the experimental treatments and calibration solutions for sweet taste.

For data collection, samples were presented at 20 ˚C in plastic soufflé cups coded with random three-digit numbers and presented in randomized order. Panelists were instructed to stir each sample for five sections with a plastic straw and then let the sample rest for 30 s prior to evaluating. All attributes were assessed on 150 mm line scales using attribute definitions provided in Table 5.2. Data were collected in Compusense® Cloud v4.7 (Guelph, Canada). A mandatory 1 min rest period was enforced between samples and deionized water, carrots, and unsalted crackers were used as palate cleansers between each sample.

Table 5.2. Sensory taste, flavor, and texture attributes with corresponding definitions and references

Attribute	Definition	Reference
Consistency	Thickness and/or density of the sample in the mouth	Weak = WMP 8% TS Moderate = WMP 12% TS Strong = WMP 20% TS
Creamy/oily	Mouthfeel associated with silky, velvety, oily, slippery, slimy, fatty, or smoothness	Threshold = WMP 11.5% TS Sub-moderate = WMP 16.6% TS
Paste	Texture associated with wallpaper paste, glue, gel, stickiness, or mucous	Weak = 0.1% CMC Moderate = 0.3% CMC
Astringency	Sensation felt in the mouth causing drying of the mouth and puckering of the cheeks	Moderate = 0.06% alum
Mouthcoating	Amount of film/product left in the mouth from the sample directly after	Weak = WMP 11.5% TS Moderate = WMP 20% TS
Sweet	Basic taste associated with sucrose in solution	Weak = 2% sucrose Moderate = 3% sucrose Strong = 4% sucrose
Salty	Basic taste associated with table salt in solution	Threshold = 0.01% salt Weak = 0.06% salt
Cream	Flavor associated with fresh New Zealand cream	Weak = SMP 10% TS Moderate = WMP 12.25% TS
Cardboard	Flavor reminiscent of wet cardboard or gelatin	Weak = 0.5% gelatine in water
Cooked	Flavor reminiscent of milk as it is heated	Weak = WMP 12.25% TS Moderate = WMP cooked 2 min
Melon/cucumber	Flavors associated with melon and cucumber, or related fruits	No reference available
Residual mouthcoating	Amount of residual film/product felt in the mouth 30 s after expectorating sample	Weak = WMP 11.5% TS Moderate = WMP 20% TS
Residual astringency	Residual aftersensation felt in the mouth causing drying of mouth and puckering of cheeks 30 s after expectorating sample	Moderate = 0.06% alum

Whole milk powder (WMP); skim milk powder (SMP); total milk solids (TS); carboxymethyl cellulose (CMC).

3.5. Statistical Analysis

Statistical analysis was performed using JMP Pro software version 13.0 (SAS, Cary, NC). Treatment effects were determined using analysis of variance (ANOVA) with Tukey's HSD (experiment–wise error set at $\alpha = 0.05$ for each response) as a post hoc multiple comparisons test to separate means where necessary. Pearson correlation coefficients were determined using multivariate analysis with pairwise comparisons; only significant values at $p < 0.05$ are displayed, and additional significance indicated at $p < 0.01$ with asterisks. Due to differences in sensory evaluation terms, separate statistical analyses were performed for aqueous, SMP, and WMP samples. Temporal data from TDS were presented as curves where attribute dominance rates were plotted as function of time (XLSTAT v18.01, Addinsoft, New York, NY). Data were smoothed within the software used to construct the curves. Statistical significance was calculated at 95% confidence using a chance rate of 11.1% based on 9 attributes.

4. RESULTS

4.1. Flow profiles

The goal was to match viscosity for each hydrocolloid pair near the shear rate in the mouth, often reported as ~50 1/s (Shama & Sherman, 1973; Wood, 1968). Three viscosity targets were selected and represent 4–6 mPa·s (low), 25–30 mPa·s (medium), and 50–60 mPa·s (high); solutions falling within each range were considered "matched viscosity." Most samples were close to these targets (Table 5.3) with the exception of medium and high viscosity carrageenan and WMP samples.

Flow profiles of aqueous solutions of λ–carrageenan (CG) and starch shown in Fig. 5.1. Flow profiles of starch samples fit a power law model ($R^2 > 98\%$; data not provided), whereas CG samples exhibited a zero-shear plateau at low shear rates with the exception of the high viscosity sample. This slight increasing trend at very low shear rates may indicate a weak fluid gel network and apparent yield stress. Starch solutions exhibited less shear thinning behavior and thus were slightly higher in viscosity across the most shear rates, most notably shear rates exceeding 1,000 1/s.

Table 5.3. Measured viscosity of hydrocolloid model systems at 50 1/s.

System	Viscosity Target	Carrageenan	Starch
Water	Low	3.4 ± 2.9	6.1 ± 1.6
Water	Medium	22.6 ± 1.9	26.9 ± 0.7
Water	High	56.5 ± 3.1	48.0 ± 2.0
SMP	Low	9.6 ± 2.8	6.4 ± 2.0
SMP	Medium	32.5 ± 3.8	26.4 ± 1.5
SMP	High	47.9 ± 2.9	50.1 ± 10.7
WMP	Low	11.1 ± 1.3	6.4 ± 3.0
WMP	Medium	41.9 ± 0.6	22.5 ± 2.4
WMP	High	74.1 ± 8.3	45.3 ± 6.6

Three viscosity targets were 4–6 mPa·s (low), 25–30 mPa·s (medium), and 50–60 mPa·s (high). Skim milk powder (SMP); whole milk powder (WMP).

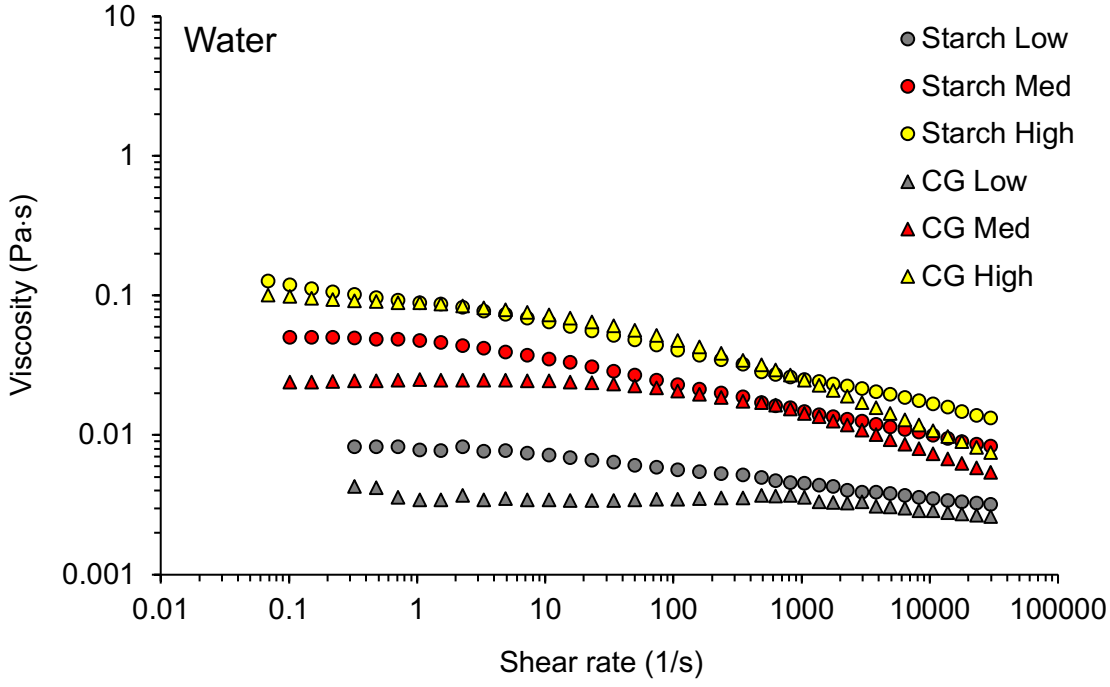

Figure 5.1. Flow profiles of starch and λ–carrageenan (CG) solutions dispersed in water at low, medium, and high viscosity targets. Couette geometry was used to determine viscosity up to 350 1/s, and parallel plate narrow gap rheology was used above 350 1/s.

Flow profiles of CG and starch dispersed in reconstituted skim milk powder (SMP) are shown in Fig. 5.2. Usage levels of each hydrocolloid were adjusted to compensate for additional solids that altered viscosity. Flow profiles of starch and SMP are similar to starch alone and still fit power law models reasonably well ($R^2 > 95\%$; data not provided). Conversely, when dispersed in SMP the CG samples exhibit a marked increase in low shear viscosity and no longer approximate a zero-shear plateau.

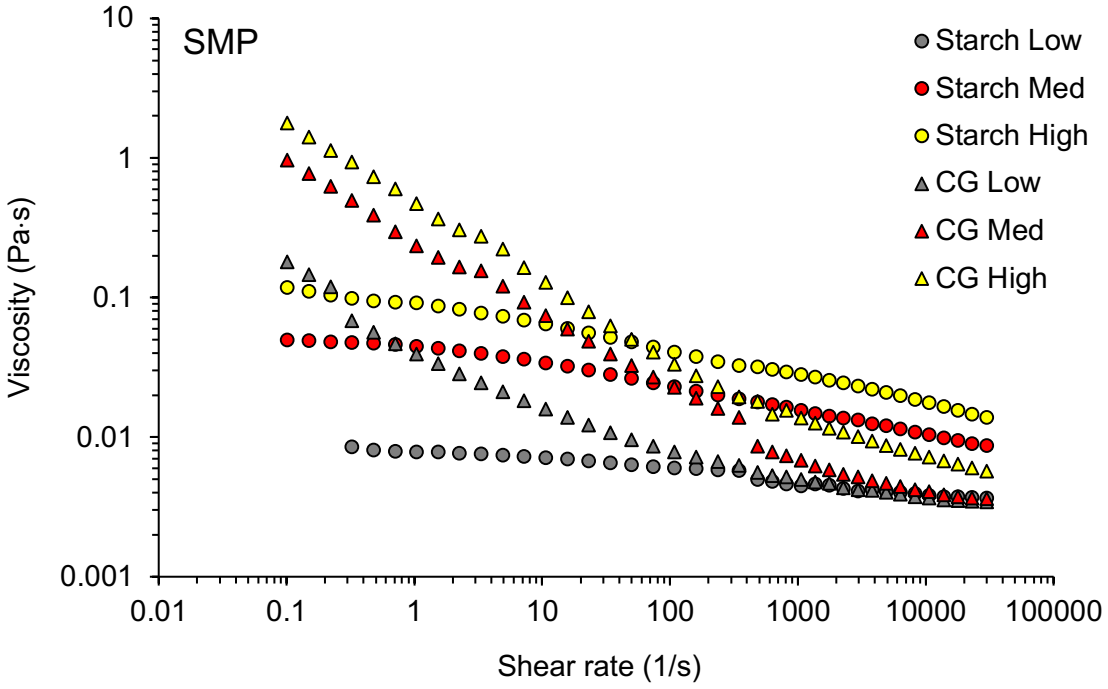

Figure 5.2. Flow profiles of starch and λ–carrageenan (CG) solutions dispersed in skim milk powder (SMP) at low, medium, and high viscosity targets. Couette geometry was used to determine viscosity up to 350 1/s, and parallel plate narrow gap rheology above 350 1/s.

Flow profiles of CG and starch dispersed in reconstituted whole milk powder (WMP) are shown in Fig. 5.3. As with SMP samples, usage levels of each ingredient were adjusted to compensate for additional solids; however, the starch samples at all three viscosity targets still exhibit slightly higher viscosity over the entire shear rate range compared to SMP samples. Flow profiles of starch and WMP are similar to starch alone and fit power law

models reasonably well (R^2 > 94%; data not provided). Conversely, CG samples exhibit a marked increase in low shear viscosity and no longer approximate a zero-shear plateau.

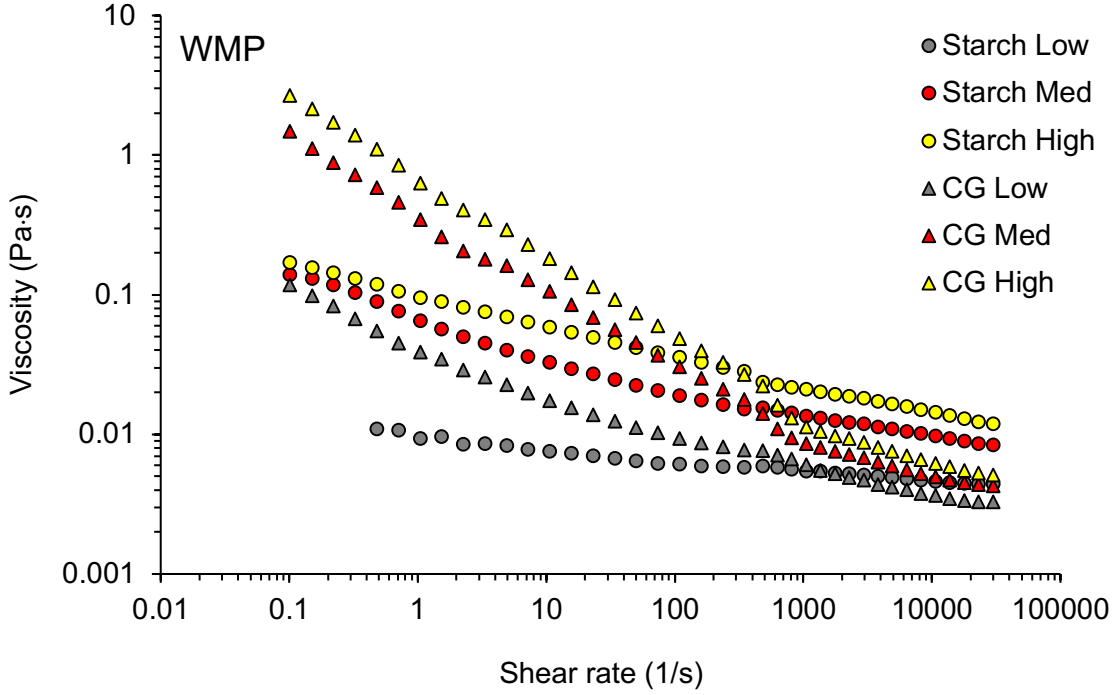

Figure 5.3. Flow profiles of starch and λ–carrageenan (CG) solutions dispersed in whole milk powder (WMP) at low, medium, and high viscosity targets. Couette geometry was used to determine viscosity up to 350 1/s, and parallel plate narrow gap rheology above 350 1/s.

4.2. Sensory properties of aqueous hydrocolloid solutions

Sensory properties of the hydrocolloid model system in water is presented in Table 5.4. Samples were differentiated in consistency, creamy/oily texture, paste, mouthcoating, cardboard flavor, and residual mouthcoating, all of which increased with increasing viscosity level. Conversely, no differences in sweet taste, salty taste, or astringency were observed based on viscosity level or hydrocolloid type. Carrageenan solutions were rated as having a higher consistency in at the high viscosity target, even though the rheological flow profiles were very similar. At the low viscosity target, starch was rated as a higher consistency, which matched sensory results.

Table 5.4. Sensory properties of aqueous hydrocolloid solutions and 3% sucrose at low, medium, and high viscosity targets.

	Carrageenan			Starch		
Attributes	**Low**	**Medium**	**High**	**Low**	**Medium**	**High**
Consistency	40.2e	86.4c	124.2a	50.0d	86.4c	109.9b
Creamy/oily	27.1e	60.5c	78.4a	35.5d	59.5c	71.4b
Paste	31.6e	67.2c	86.1a	38.0d	64.6c	77.3b
Astringency	60.1	59.1	58.9	59.8	59.2	59.7
Mouthcoating	49.2c	67.3b	77.4a	54.2c	66.7b	72.6a
Sweet	72.1	72.2	74.2	72.0	72.1	73.6
Salt	9.9	9.6	9.9	9.8	9.8	9.9
Cooked	N/A	N/A	N/A	N/A	N/A	N/A
Cardboard	21.4c	28.1ab	29.5a	22.0c	24.4bc	24.6bc
Melon/cucumber	11.1b	9.8b	10.4b	21.7a	21.6a	21.7a
Residual mouthcoating	29.2d	35.6b	40.7a	31.1c	35.6b	37.3b
Residual astringency	38.0	37.9	37.6	38.2	37.9	37.7

Letters indicate significant differences in means within rows as determined by two-way ANOVA and means separation via Tukey's HSD at $\alpha = 0.05$. Attributes are presented in the order evaluated.

These results suggest an effect of both hydrocolloid type and viscosity level on taste and texture. However, when normalized against sensory consistency (i.e., viscosity), there is no significant effect of hydrocolloid type on creamy/oily, mouthcoating, or paste (Fig. 5.4). Notably, no effect of consistency or hydrocolloid type on perceived sweet taste was observed, even across an order of magnitude increase in viscosity (3–57 mPa·s at 50 1/s). Two-way ANOVA indicates that the primary effect was due to viscosity level, and only melon/cucumber exhibited a significant effect of hydrocolloid type.

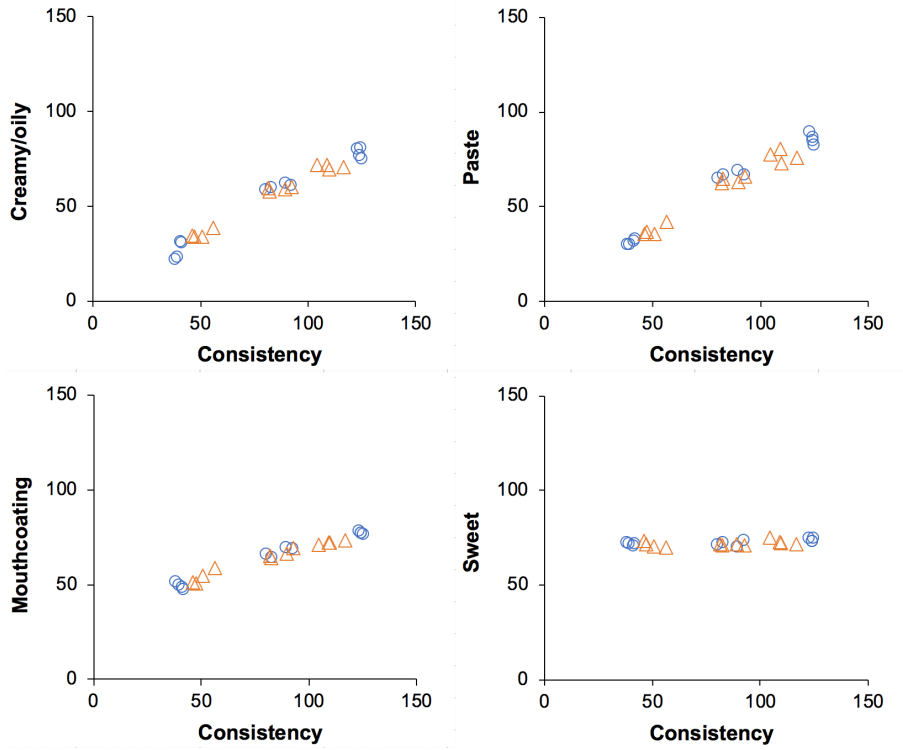

Figure 5.4. Sensory taste and texture attributes of hydrocolloid solutions normalized by sensory consistency. Starch is represented by circles and λ–carrageenan by triangles.

4.3. Sensory properties of hydrocolloid and skim milk powder solutions

Sensory properties of starch and CG dispersed in reconstituted skim milk powder is shown in Table 5. Hydrocolloid levels were reduced compared to aqueous system to account for polysaccharide-protein interactions that increased viscosity. Samples were differentiated in paste, consistency, creamy/oily texture, astringency, mouthcoating, cream flavor, residual mouthcoating, and residual astringency. These attributes were also differentiated in the aqueous model system with the exception of astringency and residual astringency. Although samples were statistically differentiated in terms of astringency, the only significant difference was between low and high viscosity starch over a small range (81–84 on a 150–point scale) so the differences among samples are very small in a practical sense. Carrageenan samples were rated as higher consistency than starch samples at all three viscosity targets, which agrees with measured viscosity at low shear rates (Fig. 5.2). Again, no differences in perception of sweet or salty taste were observed. The absolute values of

sweet taste were higher in the SMP system compared to aqueous polymers despite both being at 3% added sucrose, likely due to the presence of lactose in the powder. Interestingly, creamy texture and cream taste increased across viscosity levels for both hydrocolloids, even though the fat level remained constant (i.e., the small amount of fat present in the dried powder was constant). This indicates either panelists conflated creamy texture with creamy taste or the presence of a cross-modal effect of viscosity on creamy taste. In terms of residual attributes, highest viscosity targets showed more residual mouthcoating and astringency than the lowest viscosity targets irrespective of hydrocolloid type. However, when attributes are normalized against consistency (Fig. 5.5) there is no significant effect of hydrocolloid type; the differences in attributes are driven by viscosity level. Similar to the samples in water, no difference in sweet taste based on hydrocolloid type or viscosity level was observed despite a viscosity range of 6–50 mPa·s at 50 1/s.

Table 5.5. Sensory properties of hydrocolloid solutions in reconstituted skim milk powder with 3% sucrose at low, medium, and high viscosity targets.

	Carrageenan			Starch		
Attributes	**Low**	**Medium**	**High**	**Low**	**Medium**	**High**
Paste	34.7c	63.8ab	69.3a	21.4d	55.2b	67.7a
Consistency	63.3c	105.4ab	114.6a	52.9c	92.3b	109.2a
Creamy/oily	33.8c	55.2a	59.1a	27.2d	49.1b	57.3a
Astringency	82.2bc	82.8abc	83.3ab	81.2c	82.4ab	83.8a
Mouthcoating	51.6d	61.1bc	64.7a	46.2e	58.5c	62.2ab
Sweet	96.6	96.6	96.1	96.7	96.1	96.1
Salt	33.5	33.8	34.2	33.6	34.4	34.5
Creamy	41.3cd	41.8abc	43.0ab	40.3d	41.7bcd	43.3a
Cooked	32.7	34.5	34.5	33.9	33.7	33.0
Cardboard	26.2	27.2	26.7	26.2	27.0	26.8
Residual mouthcoating	34.8b	39.00a	39.1a	33.3b	37.6a	39.7a
Residual astringency	58.2b	59.3ab	60.0a	58.2b	59.5ab	60.0a

Letters indicate significant differences in means within rows as determined by two-way ANOVA and means separation via Tukey's HSD at $\alpha = 0.05$. Attributes are presented in the order evaluated.

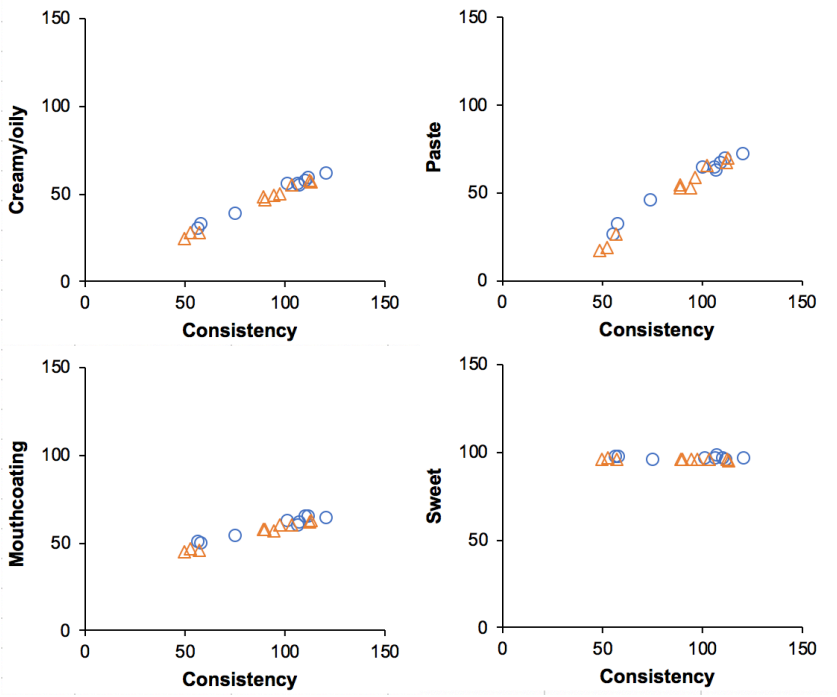

Figure 5.5. Sensory taste and texture attributes of hydrocolloid solutions in reconstituted skim milk normalized by sensory consistency. Starch is represented by circles and λ–carrageenan by triangles.

4.4. Sensory properties of hydrocolloid and whole milk powder solutions

Sensory properties of the hydrocolloid model system in reconstituted whole milk powder (WMP) is shown in Table 5.6. Samples were differentiated in terms of paste, consistency, creamy/oily, mouthcoating and residual mouthcoating, although the significant differences were generally only between the lowest and highest viscosity levels. No significant hydrocolloid effect was observed across all viscosity levels although CG samples tended to be higher in perceived paste, consistency, creamy/oily, and mouthcoating. Compared to SMP samples, WMP samples were reported as slightly less sweet, salty, and astringent. The decrease in salt and sweet taste can both be attributed to the lower lactose and mineral content of WMP, whereas the higher fat in WMP would likely decrease astringency and increase creamy texture and taste. Normalized by consistency (Fig. 5.6), the trends are the same as with SMP: no effect of hydrocolloid type and the main effect is viscosity level. Interestingly, no effect on sweet taste was observed despite viscosity ranging from 6–74 mPa·s at 50 1/s.

Table 5.6. Sensory properties of hydrocolloid solutions in reconstituted whole milk powder with 3% sucrose at low, medium, and high viscosity targets.

	Carrageenan			Starch		
Attributes	**Low**	**Medium**	**High**	**Low**	**Medium**	**High**
Paste	46.7cd	68.5ab	78.8a	31.6d	55.7bc	68.4ab
Consistency	78.1cd	115.3ab	131.7a	65.6d	92.8bc	116.0ab
Creamy/oily	48.0bc	58.6ab	63.3a	39.6c	52.5ab	58.6ab
Astringency	74.5	75.1	75.8	75.2	75.2	75.5
Mouthcoating	67.6b	71.7ab	76.9a	65.5b	69.2ab	72.9ab
Sweet	95.6	95.8	96.0	95.6	95.7	95.2
Salt	30.5	31.0	31.8	30.6	31.6	31.0
Creamy	67.1	68.4	68.4	66.7	67.4	68.2
Cooked	42.0	42.4	42.7	42.9	42.9	43.2
Cardboard	0	0	0	0	0	0
Residual mouthcoating	39.4bc	42.0abc	43.4a	38.4c	39.6bc	42.6ab
Residual astringency	51.0	53.0	53.2	51.1	51.2	52.0

Letters indicate significant differences in means within rows as determined by two-way ANOVA and means separation via Tukey's HSD at $\alpha = 0.05$. Attributes are presented in the order evaluated.

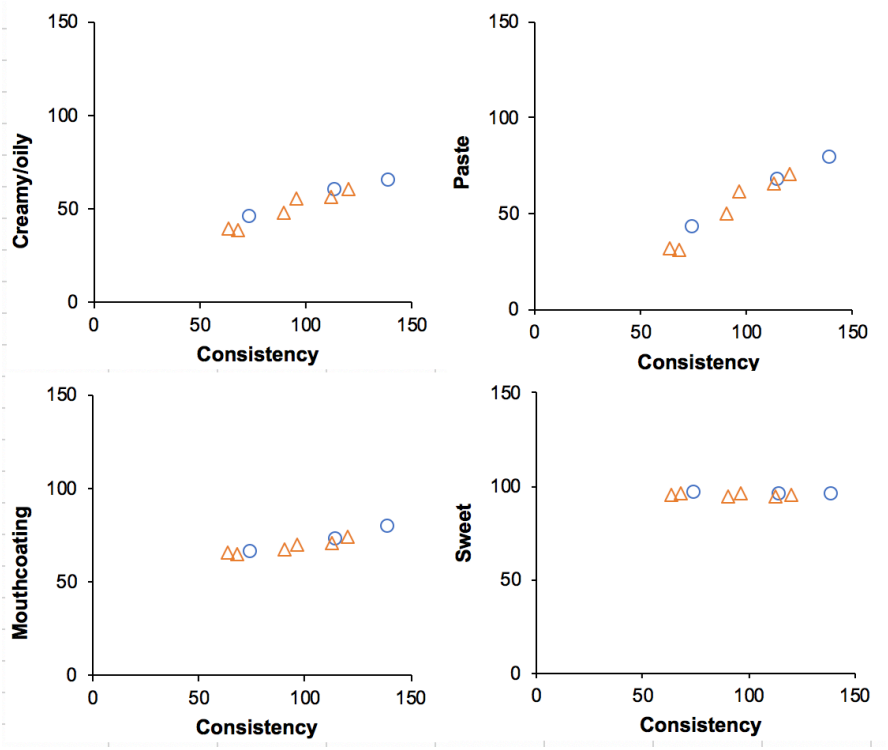

Figure 5.6. Sensory taste and texture attributes of hydrocolloid solutions in reconstituted whole milk normalized by sensory consistency. Starch is represented by circles and λ–carrageenan by triangles.

4.5. Temporal dominance of sensory attributes

Temporal progression of sensory attributes is shown in Fig. 5.7. Six samples were selected to allow comparisons of viscosity, hydrocolloid type, and milk powder type. Preliminary work showed that sweetness was overwhelmingly dominant across all samples; therefore, to fully explore secondary attributes pre- and post-expectoration the sweet taste selection was removed. Skim milk powder without hydrocolloids was characterized by sweet aromatic and cooked flavors followed by post-expectoration astringency. The panel did not come to a consensus for samples with starch or CG, which showed dominant thickness, creaminess, cooked flavor, and sweet aromatic flavor transitioning to mouthcoating and astringency. Whole milk powder samples without hydrocolloids were similar to SMP with the addition of pre-expectoration dominant creaminess. Starch samples were also similar in both SMP and WMP. However, CG in WMP exhibited mainly dominant creaminess leading to sweet aromatic flavor, mouthcoating, and astringency post-expectoration.

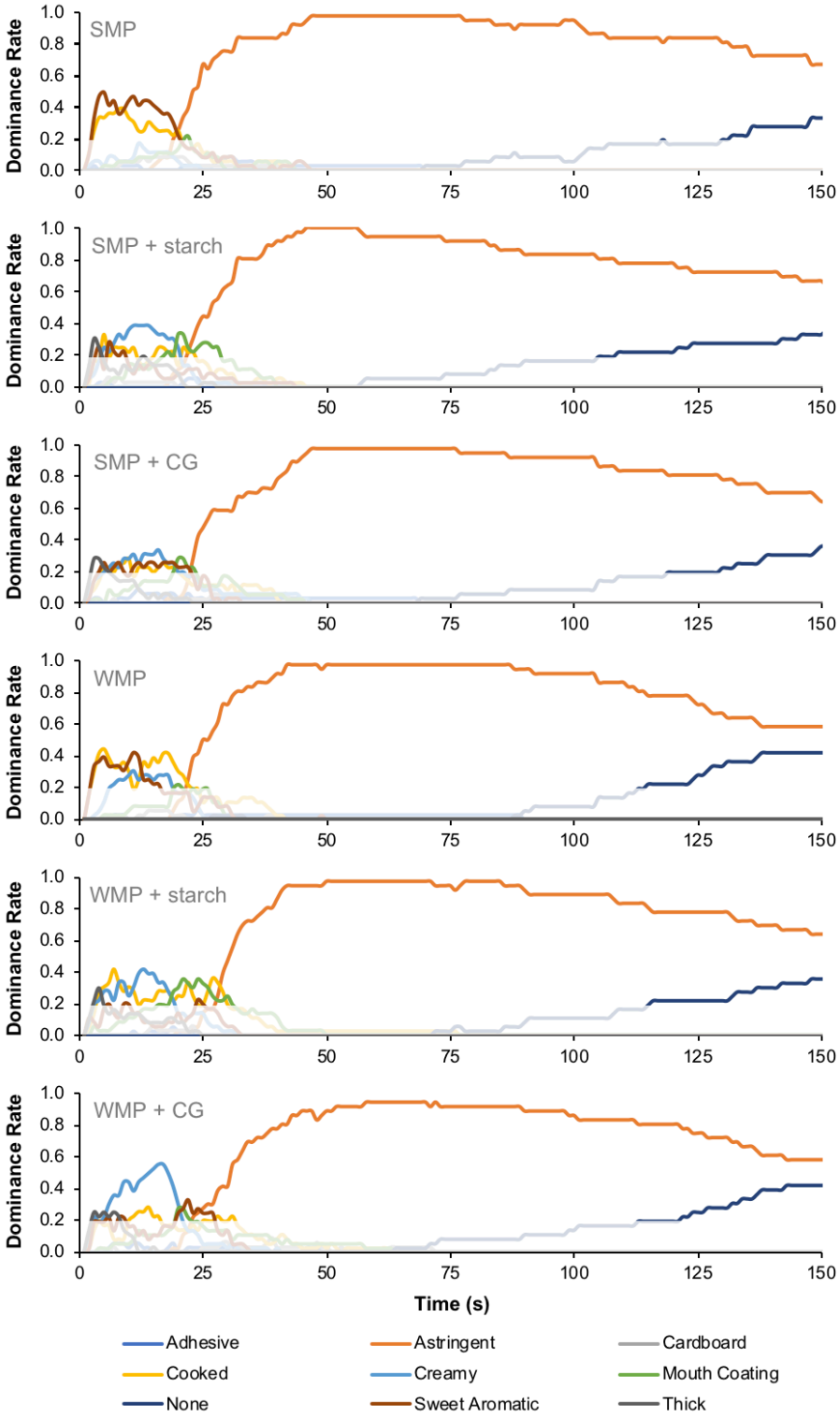

Figure 5.7. Temporal profiles of hydrocolloids in reconstituted skim milk and whole milk powders. Data are faded below the significance level of 0.197 based on $\alpha = 0.05$. Whole milk powder (WMP); skim milk powder (SMP); carrageenan (CG).

Table 5.7. Correlations among sensory and rheological attributes of hydrocolloid solutions dispersed in reconstituted whole milk powder.

	Paste	Consistency	Creamy/Oily	Astringency	Mouthcoating	Sweet	Salt	Creamy	Cooked	Residual mouthcoating	Residual astringency	Visc 0.5 s^{-1}	Visc 50 s^{-1}	Visc 30,000 s^{-1}
Paste	1.00	0.98*	1.00*		0.94*			0.88*		0.93*	0.77		0.94*	
Consistency		1.00	0.97*	0.78	0.97*			0.84*		0.95*	0.77	0.76	0.98*	
Creamy/Oily			1.00		0.94*			0.89*		0.93*	0.81		0.92*	
Astringency				1.00	0.81					0.85	0.72		0.80	
Mouthcoating					1.00			0.75		0.96*	0.85*	0.84	0.99*	
Sweet						1.00								
Salt							1.00							
Creamy								1.00		0.85	0.72		0.77	
Cooked									1.00					
Residual mouthcoating										1.00	0.90*	0.75	0.95*	
Residual astringency											1.00	0.76	0.79	
Visc 0.5 s^{-1}												1.00	0.84*	
Visc 50 s^{-1}													1.00	
Visc 30000 s^{-1}														1.00

Only significant values shown ($p < 0.05$), asterisk indicates $p < 0.001$.

4.6. Correlations among sensory attributes and viscosity

Correlations among sensory and rheological attributes of SMP and WMP model systems are shown in Tables 5.7 and 5.8, respectively. Attributes associated with viscosity such as paste, consistency, creamy, and mouthcoating tended to be strongly correlated in both SMP and WMP model systems. These same attributes correlated strongly with viscosity at 50 1/s. Viscosity at 30,000 1/s did not correlate with any attributes in either model system. Sweet and salty taste were not correlated with any attributes. Residual astringency after expectoration correlated with paste, consistency, creamy, astringency, and mouthcoating but only in the WMP system.

Table 5.8. Correlations among sensory and rheological attributes of hydrocolloid solutions dispersed in reconstituted skim milk powder.

	Paste	Consistency	Creamy oily	Astringency	Mouthcoating	Sweet	Salt	Creamy	Cooked	Cardboard	Residual mouthcoating	Residual astringency	Visc 0.5 s^{-1}	Visc 50 s^{-1}	Visc 30,000 s^{-1}
Paste	1.00	0.99*	0.99*		0.98*						0.96*			0.97*	
Consistency		1.00	0.99*		0.98*			0.82			0.95*			0.97*	
Creamy oily			1.00		0.98*			0.82			0.95*			0.96*	
Astringency				1.00				0.88						0.99*	
Mouthcoating					1.00						0.95*			0.96*	
Sweet						1.00									
Salt							1.00								
Creamy								1.00						0.95*	
Cooked									1.00						
Cardboard										1.00					
Residual mouthcoating											1.00			0.92*	
Residual astringency												1.00		0.83	
Visc 0.5 s^{-1}													1.00		
Visc 50 s^{-1}														1.00	
Visc 30000 s^{-1}															1.00

Only significant values shown ($p < 0.05$), asterisk indicates $p < 0.001$.

5. DISCUSSION

Beverages can contain a variety of ingredients that interact to produce structural complexity across many different length scales. The inclusion of hydrocolloids to change viscosity, enhance stability, or to alter mouthfeel or other textural properties is common. However, many of these ingredients also interact with the oral mucosa and can influence product properties during consumption. Previous studies have suggested that mucoadhesion may play a role in modulating taste and texture perception (Cook et al., 2017; Cook, Woods, et al., 2018). Therefore, understanding how mucoadhesion and texture-taste interactions modulate taste perception is important for the design of foods with targeted health or functional applications.

The initial objective of this study was to evaluate the role of mucoadhesion in taste perception by selecting a range of hydrocolloids that exhibit differences in binding affinity to mucins. A milk protein concentrate model system with 3% sucrose was developed based on the results of Chapter 3. Hydrocolloid usage levels were adjusted in order to match viscosity near 6–8 mPa·s at 50 1/s (Appendix A). Confocal microscopy showed a range of microstructures depending on type of hydrocolloid, including fluid gels, colloidal dispersions, homogenous dispersions, and micro-phase separated systems (Appendix A). However, preliminary sensory data confirmed that at this viscosity level, hydrocolloid type did not alter perceived thickness or sweet taste.

Based on this preliminary study, alternative model systems were developed around three factors: viscosity level, hydrocolloid type, and dispersion media. Tapioca starch and λ–carrageenan (CG) were selected as ingredients as previous reports suggest that anionic polysaccharides bind mucins more strongly than starch-based systems (Cook et al., 2018). Despite both contributing to bulk viscosity, they vary in microstructure where starch is a granular dispersion depending on degree of gelatinization and CG is a random coil polysaccharide. Starch solutions are thought to mix more efficiently with saliva than polymeric hydrocolloids, which may contribute to decreased taste suppression compared to polymeric systems (Ferry et al., 2006). Differences in how the products break up due to shear forces during oral processing and activity of salivary amylase to hydrolyze starch and reduce viscosity should also be considered (Desse, Mitchell, Wolf, & Budtova, 2011). Considering starch and CG have different flow profiles, viscosity cannot be matched *per se*, therefore samples were formulated to match viscosity near 50–100 1/s as low shear rate ranges have previously been reported to correlate strongly with taste perception (Aubert et al., 2016; He et al., 2016). In the present study, viscosity at 0.5, 50, and 30,000 1/s were selected for analysis, and viscosity at 50 1/s correlated with more taste and texture attributes than 0.5 1/s. Viscosity at 30,000 1/s did not correlate with any taste or texture attributes. Additionally, attributes related to thickness (e.g., paste, consistency, creamy/oily, mouthcoating) tended to be strongly correlated in both SMP and WMP systems.

In water, viscosity level was the significant treatment effect and only melon/cucumber flavor exhibited any significant hydrocolloid type effect. This flavor is likely an inherent quality of the ingredients. Moreover, the usage levels were higher than

normal amounts because no other ingredients were present, so the high hydrocolloid levels would likely exacerbate off flavors compared to usage levels in milk systems. Melon/cucumber was not detectable in the milk-based systems. Similar effects were observed for the milk protein samples where viscosity level was a much more significant effect than hydrocolloid type.

The presence of fat may change taste and texture directly, where it acts as a solvent for lipophilic flavor compounds, or indirectly, where it may alter underlying food structure (Arancibia, Castro, Jublot, Costell, & Bayarri, 2015; Malone, Appelqvist, & Norton, 2003; Panouillé, Saint-Eve, de Loubens, Déléris, & Souchon, 2011). In the present study, SMP and WMP differed primarily in fat content with slight differences in mineral and lactose levels. The WMP samples were perceived to be higher in paste, consistency, creamy, mouthcoating, and lower in astringency than SMP samples. Many of these are associated with viscosity; however, the viscosities were very similar across all shear rate ranges. This points towards lubrication playing a role in creaminess perception or potential cross-modal interactions among texture attributes (Chojnicka-Paszun, de Jongh, & De Kruif, 2012; de Wijk & Prinz, 2007).

The notion that viscosity-based taste suppression is independent of hydrocolloid type would be attractive to food manufacturers. Research findings in this area are mixed; some investigations have reported no hydrocolloid type effect (Cook et al., 2003; Hollowood, Linforth, & Taylor, 2002; Lethuaut, Brossard, Rousseau, Bousseau, & Genot, 2003), while others have reported a clear effect of hydrocolloid type (Christensen, 1980; Pangborn et al., 1973). Ferry et al. reported lower flavor and taste in model savory sauces thickened with hydroxymethyl propyl cellulose (HPMC) rather than maize starch under the hypothesis that lower mixing efficiency with saliva and a lack of contribution of α–amylase to decrease viscosity in mouth (Ferry et al., 2006). However, significant taste suppression wasn't realized until a viscosity of nearly 300 mPa·s for HPMC and almost 400 mPa·s for starch. Moreover, the viscosity of these samples ranged from 80–480 mPa·s (at 50 1/s), which would be considerably higher than most beverages. This suggests that at viscosities typical of beverages there may not be a strong effect of hydrocolloid type on taste or texture attributes. One could argue that considering taste suppression occurs at polymer concentrations above c*, that commercial usage levels are below c* or that this mechanism doesn't apply to

complex colloidal systems (Baines & Morris, 1987; Cook et al., 2002). Additionally, considering starch has been reported to be less mucoadhesive than anionic polymers, this may indicate that greater viscosity levels or higher hydrocolloid usage levels would be required to realize a noticeable sensory difference. Arancibia et al. reported an effect of hydrocolloid type on texture, flavor, and taste perception of semisolid dairy matrices but these products are much more viscous and may have other oral processing conditions to consider (Arancibia et al., 2015). Similar results were found in Ch. 3, where several orders of magnitude increase in viscosity was required to suppress taste in a polymeric system, and in a milk protein system there was no effect. Another possibility is that the type of hydrocolloid matters depending on sucrose level. Lethaut et al. evaluated semisolids thickened with different types of carrageenan, and an observable hydrocolloid type effect depended on the sucrose load (Lethuaut et al., 2003).

Mucoadhesion has also been implicated in the oral-tactile sensation of astringency (Bull et al., 2017). In the present study, astringency correlated with mouthcoating, viscosity (50 1/s), and residual mouthcoating in the WMP system. Astringency by definition is a lingering sensation so it is intuitive that mouthcoating attributes or others that facilitate binding would enhance this sensation (Courregelongue, Schlich, & Noble, 1999). Astringency of dairy products has been reported due to whey proteins ability to bind and precipitate salivary proteins (Vardhanabhuti, Kelly, Luck, Drake, & Foegeding, 2010; Vingerhoeds, Silletti, de Groot, Schipper, & van Aken, 2009). However, both caseins and β–lactoglobulin have been shown to be mucoadhesive indicating that they both may lead to the perception of astringency (Withers et al., 2013). In this study, astringency was reported as higher in SMP samples despite viscosity being slightly lower. This is likely due to the lack of fat because fat suppresses astringency (des Gachons et al., 2012), so there may be some trade-off between viscosity and fat level in terms of modulating astringency.

It is less known how mucoadhesive hydrocolloids modify temporal taste and texture perception. Cook et al. used progressive profiling of attribute intensity over time and reported that CMC was more mouthcoating and adhesive than starch at matched dynamic viscosity at 50 rad/s (Cook, Woods, et al., 2018). We selected temporal dominance of sensations (TDS) as a tool to qualitatively assess a greater number of attributes compared to progressive profiling or time intensity analysis, but at the expense of quantitative data. The primary

differences were between the type of powder used (i.e., SMP or WMP). Astringency onset and persistence did not seem to change based on hydrocolloid type or presence of fat in WMP.

6. CONCLUSIONS

Results of this study indicate that despite reported differences in mucoadhesion between a random coil polysaccharide and a modified starch, there are few effects on perceived taste and texture in dairy-based matrices. The primary effect was due to hydrocolloid addition increasing viscosity and associated texture terms. This trend held true for a range of viscosity levels representing thin and thick beverages. Despite the range in viscosity, sensory perception of basic tastes (sweet and salty) and flavor (cardboard and cooked) were not altered. Only viscosity at 50 1/s correlated with all texture terms in both SMP and WMP model systems, whereas viscosity at 0.5 1/s only correlated when fat was present. Samples did not exhibit marked differences in the temporal progression of dominant sensory attributes. Viscosity appears to be the most important attribute in altering texture, but significant across the viscosity range tested in the study there was no effect on perceived taste.

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CHAPTER 6. CARAMEL AS A MODEL SYSTEM FOR EVALUATING THE ROLES OF MECHANICAL PROPERTIES AND ORAL PROCESSING ON SENSORY PERCEPTION OF TEXTURE

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Caramel as a Model System for Evaluating the Roles of Mechanical Properties and Oral Processing on Sensory Perception of Texture

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Abstract: Food formulation can have a significant impact on texture perception during oral processing. We hypothesized that slight modifications to caramel formulations would significantly alter mechanical and masticatory parameters, which can be used to explain differences in texture perception. A multidisciplinary approach was applied by evaluating relationships among mechanical properties, sensory texture, and oral processing. Caramels were utilized as a highly adhesive and cohesive model system and the formulation was adjusted to generate distinct differences in sensory hardness and adhesiveness. Descriptive analysis was used to determine sensory texture, and mechanical properties were evaluated by oscillatory rheology, creep recovery, and pressure sensitive tack measurements. Oral processing was measured by determining activity of anterior temporalis and masseter muscles via electromyography and tracking jaw movement during chewing. The substitution of agar or gelatin for corn syrup at 0.6% w/w of the total formulation resulted in increased sensory hardness and decreased adhesiveness. Creep recovery and pressure sensitive tack testing were more effective at differentiating among treatments than oscillatory rheology. Hardness correlated inversely with creep compliance, and both stickiness and tooth adhesiveness correlated with pressure sensitive adhesive force. Harder samples, despite being less adhesive, were associated with increased muscle activity and jaw movement during mastication. Tooth packing, not linked with any mechanical property, correlated with altered jaw movement. The combination of material properties and oral processing parameters were able to explain all sensory texture differences in a highly adhesive food.

Keywords: adhesiveness, confections, cohesiveness, electromyography, oral processing, texture

Practical Applications: Food structure, texture, and oral processing are important parameters for the assessment of food quality, satiation and satiety, and acceptance of foods within certain population segments. Understanding how oral processing is altered due to compositional changes will provide a better understanding of texture perception during oral processing. This study provides insight into the relationships among mechanical properties, sensory perception, and oral processing, which can be used to better understand the mechanisms responsible for the adhesive and cohesive properties of foods and help design textures for specific food applications.

Introduction

Texture is an important attribute of food quality that originates from food structure; however, the mechanisms responsible for texture perception are not fully understood. Although texture is difficult to define due to the inherent complexity of the term, many agree that "*texture is the sensory and functional manifestation of the structural, mechanical and surface properties of foods detected through the senses of vision, hearing, touch and kinesthetics*" (Szczesniak 2002). Texture is an important driver of sensory perception and consumer acceptance of foods (Guinard and Mazzucchelli 1996; Szczesniak 2002) and, more recently, has been demonstrated to play a role in satiation and satiety (de Graaf and Kok 2010). The pathway of food breakdown during oral processing is one of many variables that influence the perception of texture. In order to explain how food structure is transformed into the perception of specific textural properties, a better understanding of the relationships among initial structural properties, structural transformations during oral processing, and their connection to sensory perception is needed (Chen 2009; Foster and others 2011; Pascua and others 2013).

Food texture investigations can be grouped into 3 general areas: forming and stabilizing food structures, breakdown of structure during oral processing, and sensory perception of texture during oral processing. Physical properties are related to structural elements, thus providing insight into how manipulation of food structure can produce specific textural properties. Foods can be considered materials and amenable to measuring physical properties through rheological and fracture mechanics tests (Vincent 2004). Thus, the most common approach for investigating structure–texture relationships is to perform mechanical and sensory testing on the same food and look for correlations (Foegeding and Drake 2007; Stieger and van de Velde 2013). Correlations depend on how the sensory term is assessed. Sensory terms, such as first bite or chew hardness, are evaluated by assessing intra-oral force–deformation relationships. This is operationally similar to mechanical tests determining force–deformation properties. Therefore, they are logically correlated with mechanical properties, especially with hard-solid foods that show a clear

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fracture (Kim and others 2012). In these cases, both human and machine are essentially manipulating the food in the same way. In contrast, more complex terms associated with later stages of oral processing, also called "chew down" terms, may not be directly associated with mechanical properties of the starting food material. This could be because the appropriate material property is not being measured, or because sensory terms are assessed after the material has transformed during oral processing, such as an increase in moisture content or breakdown of constituent molecules (de Wijk and others 2004).

The ability to fine-tune microstructure often drives choice of foods and model foods used to evaluate texture. For example, the links among physical properties, structure, sensory texture, and oral processing have been evaluated using soft-solid biopolymer gels and cheese (Çakir and others 2012b; Ishihara and others 2013; Melito and others 2013). Foster and others (2006) used elastic and plastic confectionary model systems to evaluate the effects of rheological parameters on oral processing. The results of these studies indicate that certain oral processing parameters, namely muscle activity and jaw movement, can elucidate the connection between physical properties and sensory texture. The present study builds on the foundation of these studies by utilizing a model system that is both adhesive and cohesive to gain a better understanding of what the human is evaluating and perceiving during mastication.

Caramel structure can be explained based on colloidal and polymer models. From a colloidal perspective, emulsified fat is dispersed in a continuous phase consisting primarily of carbohydrate (for example, polymers, oligomers, mono-, and disaccharides) but can also contain protein. The low water content—typically 5% to 10%—and high phase volume of carbohydrates can result in an amorphous structure that fits polymer models. Caramel also possesses unique properties that are important for an understanding of texture perception. Unlike many soft-solid or hard-solid foods, caramels, and other confections are often highly cohesive, and do not readily fracture during mastication. Therefore, the process of comminution is limited, and oral processing primarily involves material transformations and removal from oral surfaces, which continue until the bolus can be safely swallowed (Brown and others 1998).

The objective of this study was to explain sensory texture terms by determining how oral processing parameters change in response to small compositional differences in caramels. We hypothesize that oral processing parameters are adjusted to manipulate food structure, with different structures possessing different oral processing "fingerprints." A proportion of corn syrup in a standard caramel formula was replaced with agar or gelatin to generate 3 distinct samples exhibiting differences in sensory hardness and adhesiveness. Mechanical properties, sensory texture, and oral processing of the caramels were evaluated, and relationships among the 3 were investigated. The results of this study indicate that small compositional differences alter textural properties, and sensory texture can be explained based on mechanical properties of the starting material and oral processing patterns.

Materials and Methods

Caramel formulation and standardization

Anhydrous milk fat with a reported melting temperature of 37 °C was provided by Fonterra (Auckland, New Zealand). Gelatin was provided by Vyse (275 Bloom Type A, Schiller Park, Ill., U.S.A.) and agar was provided by TIC Gums (Agar Agar 100, White Marsh, Md., U.S.A.). Granulated cane sugar (Domino®, Domino Foods Inc., Iselin, N.J., U.S.A.), corn syrup reported as 42 DE by manufacturer (Karo® Light; ACH Food Companies, Inc., Cordova, 'Tenn., U.S.A.) and sweetened condensed skim milk (Eagle Brand®, J.M. Smucker Company; Orrville, Ohio, U.S.A.) were purchased at a local grocery. Three caramel samples were prepared following the formulations in Table 1. The standard caramel of Steiner and others (2003) was modified as a caramel "control," and test caramels were formulated by replacing equal parts corn syrup (by weight) with 0.6% w/w agar or gelatin. Caramels were prepared in 600 g batches according to the method of Steiner and others (2003) with several modifications. Briefly, a surface plate heater was set to 180 °C to melt anhydrous milk fat in a 1.5 L stainless steel aluminum core pan (All-Clad, Metal Crafters Inc., Canonsburg, Pa., U.S.A.) before adding sucrose, corn syrup, sweetened condensed skim milk and deionized water. The mixture was mixed at 315 rpm with an overhead stirrer (Heidolph Instruments, Schwabach, Germany) for 20 min and a thermocouple was used to monitor temperature during heating. The plate heater was then increased to 375 °C and the stirring speed was reduced to 200 rpm until the mixture reached 119 °C. The caramel was mixed by hand for 5 s before being poured into a rectangular steel mold (30 cm × 14 cm × 0.6 cm) sprayed with food-grade silicon. Each batch was cooled on a marble slab for 60 min and vacuum packaged for storage to limit moisture loss. Caramels were stored for at least 24 h and no more than 7 d prior to sensory or mechanical analysis.

Table 1–Caramel formulations by weight percentage (%).

Ingredient	Control	Agar	Gelatin
Water	14.9	14.9	14.9
Sucrose	30.4	30.4	30.4
Light corn syrup (42 DE)	24.7	24.1	24.1
SCSM[a]	20.0	20.0	20.0
Anhydrous milk fat	10.0	10.0	10.0
Gelatin	0.0	0.0	0.6
Agar	0.0	0.6	0.0
Total (%)	100.0	100.0	100.0

Formulations are modified from Steiner and others (2003).
[a]SCSM: sweetened condensed skim milk.

Caramels were standardized by both moisture content and glass transition temperature. Moisture content was measured via Karl Fischer Titration on a 701 KF Titrino (Metrohm, Herisau, Switzerland) using Hydranal Composite-5 (Sigma-Aldrich, St. Louis, Mo., U.S.A.) as the titrating solution and 100% methanol as the solvent. Water heated to 60 °C was circulated around the titration vessel to facilitate solvation of the caramel samples. The titrating solution was standardized in triplicate using 20 μL measures of deionized water. Samples (~0.25 g) were added to the titration vessel and stirred with a magnetic stir bar for 600 s until completely dissolved. The glass transition temperature (T_g) of each caramel sample was measured using a DSC 7 (Perkin Elmer, Waltham, Mass., U.S.A.) Differential Scanning Calorimeter. Approximately 90 mg of caramel was placed in an aluminum pan and heated from –30 to 60 °C at a rate of 5 °C/min. Nitrogen gas purged the DSC at a rate of 20 mL/min. The heat flow was analyzed using the accompanying software (Pyris v5.0, Perkin Elmer, Waltham, Mass., U.S.A.) to determine the midpoint of the T_g of each sample. Measurements for both tests were performed in triplicate.

Descriptive sensory analysis

Sensory analysis was performed via the Spectrum™ method of descriptive analysis for generation of qualitative and quantitative data (Meilgaard and others 1991). Seven ($n = 7$) highly trained panelists (all female, ages 50 to 70) were selected based on experience and availability, each with over 500 h of descriptive analysis experience and approximately 250 h of experience specific to food texture. Two preliminary training and discussion sessions were held to familiarize panelists with the product specific lexicon of Steiner and others (2003) provided in Table 2. Hardness was evaluated at first bite, and the remaining attributes were evaluated in the order listed in Table 2 as the panelists chewed naturally. Fourteen different commercially available products were provided during training to represent a range of intensities for the attributes and to establish calibration standards and scale anchors. Texture attributes were evaluated on a 15-point product specific scale with 15 representing "very" and 0 representing "not at all." Caramel samples were cut into cuboids (2.5 cm × 2.5 cm × 0.6 cm) 2 h prior to analysis. Each panelist was presented with 3 replications of each sample separated by parchment paper in a lidded 2 oz clear plastic soufflé cup. Sample cups were labeled with a random 3-digit code and the order of samples was randomized. Distilled, deionized water and carrots were used as palate cleansers between samples.

Rheological analysis

A controlled stress rheometer (Anton Paar MCR 302, Graz, Austria) equipped with 25-mm dia. parallel plates was used to conduct rheological analyses at room temperature (22 ± 1 °C). Sandpaper (60 grit, Ali Industries, Fairborn, Ohio, U.S.A.) was affixed to both plates with double-sided adhesive tape (3M, St. Paul, Minn., U.S.A.) to reduce slipping during measurements. Caramel samples were cut into disks (20-mm dia., 5 mm height) and compressed to a 2 mm gap between plates. Samples were allowed to equilibrate to a 0.1 N normal force prior to measurements. Stress sweeps from 0.01 to 15 kPa were performed in oscillatory shear mode to resolve the linear viscoelastic region (LVR), and frequency sweeps from 0.01 to 100 Hz were conducted within the LVR to determine the mechanical spectra. Creep recovery tests were also conducted as compliance (J) has previously been associated with sensory texture terms (Brown and others 2003). Using the aforementioned geometry, a constant 1 kPa stress was instantaneously applied and held for 60 s for the creep portion, and samples were then allowed to recover for 300 s. Initial compliance (J_0) was the first value after the stress was applied. Maximum compliance (J_{max}) was the maximum value reached during the creep portion. Retardation time (λ_{ret}) was the amount of time for the sample to reach 63.2% of J_{max} (Steffe 1996). Percentage recovery (crp) was the percent of compliance recovered after relaxation (Eq. 1), where J_r is the compliance nadir after relaxation.

$$crp = \frac{J_{max} - J_r}{J_{max}} \times 100 \quad (1)$$

Pressure sensitive tack

Mechanical adhesion was determined using a universal testing machine (Instron 5565, Norwood, Mass., U.S.A.) equipped with a flat 56-mm dia. stainless steel probe. Samples were cut into discs (20-mm dia., 7 mm height) and placed on the stainless steel platform below the probe. Each testing cycle involved 3 steps (compression, hold, decompression) applied once to each sample. The probe was lowered to the surface of the sample, which was then compressed to 20 N at a rate of 2 N/s. The load was held for 10 s and the probe was removed at a rate of 1.5 mm/s. Tack force is defined as the maximum force recorded during decompression.

Oral processing

Nine ($n = 9$) untrained subjects (8 females, 1 male, ages 22 to 30) participated in the oral processing study and gave voluntary informed consent. The study was conducted in accordance with the North Carolina State University Institutional Review Board for the Protection of Human Subjects in Research guidelines (IRB # 3327). All subjects presented Class I type molar occlusion and had complete dentition with the exception of third molars or wisdom teeth. Subjects attended a preliminary session to acclimate to the environment, recording systems, and sample type. Prior to measurements, maximum jaw motions were measured in vertical, anterior posterior and medial lateral dimensions to ensure proper range of jaw motion. Only participants with at least 40 mm vertical and 7 mm medial lateral range of jaw movement were allowed to participate.

Jaw movement and muscle activity of all subjects were recorded according to the method of Çakir and others (2012a) with several modifications. In short, electromyographic (EMG) activities of the left and right superficial masseter and anterior temporalis muscles were recorded via attached surface electrodes (BioResearch Inc., Milwaukee, Wis., U.S.A.). An additional electrode was placed on the shoulder to eliminate background noise. Data were recorded with a BioEMG II electromyograph (BioResearch Inc.) at a sampling rate of 5000 Hz. Electrodes were connected to a single channel amplifier to allow for data collection during mastication.

Three-dimensional jaw movements were collected simultaneously using a jaw tracking device (JT–3D, BioResearch Inc.). A magnet was affixed to the frontal incisors with adhesive (Conva-Tec, Bristol–Myers Squibb Co., Skillman, N.J., U.S.A.), allowing for detection of jaw movement in vertical, medial lateral and anterior posterior directions. Caramel samples were cut into cuboids (2.5 cm × 2.5 cm × 0.6 cm) as per samples used in sensory analysis. Panelists were instructed to begin with the sample on the middle of their tongue and teeth at occlusion. The researcher then instructed the panelist to chew in a habitual manner and fully consume the sample. Data were collected from each subject in multiple sessions with 2 replicates of each treatment presented in random order.

Data analysis was conducted using an in-house program written in LabView (National Instruments, Austin, Tex., U.S.A.). Raw EMG signals were integrated using a root mean square function over the entire chewing sequence. Muscle activity values were scaled within-panelists for both peak activity and area under the curve; each chewing cycle was normalized against the largest value per each individual's recording session, resulting in values ranging from 0 to 1. This was done to control for variations associated with electrode attachment between replications. Values from right and left sides were combined and reported as total muscle activity. Some chewing cycles contained intermittent jaw movements without occlusion; these values were excluded from calculations. Definitions of oral processing terms are presented in Table S1.

Statistical analysis

Statistical differences among treatment means were determined using the one-way analysis of variance feature with Tukey's honest significant difference test in JMP 12.0 (SAS, Cary, N.C., U.S.A.).

Table 2–Sensory term definitions provided to sensory panelists[a].

Sensory term	Description
Hardness	Amount of force required to bite completely through the sample with the molars
Cohesiveness of mass	Degree to which the sample holds together in a mass at the specified number of chews[b]
Stickiness	Degree to which the product adheres to any mouth surface (including teeth) while chewing
Toothpacking	Amount of product packed in and between molars
Tooth adhesiveness	Amount of force required to remove product that adheres to teeth

[a]Terms were evaluated by panelists in the order presented from top to bottom.
[b]Cohesiveness was evaluated after 13 to 15 chews and 30 chews, which corresponded to approximately 25% and 50% of the total number of chews required to consume each sample, respectively.

Table 3–Mean texture values of caramel samples on a 15-point scale as determined by descriptive sensory evaluation.

Sensory term	Control	Agar	Gelatin	F ratio	*P*-value
Hardness	6.3[c]	10.4[a]	8.8[b]	328	<0.0001
Cohesiveness (13–15)[1]	12.9[a]	12.0[b]	13.2[a]	19.8	<0.0001
Stickiness	13.1[a]	10.5[c]	11.8[b]	92.1	<0.0001
Cohesiveness (30)[1]	12.6	12.7	12.3	NS	
Toothpacking	8.0[a]	7.1[b]	7.1[b]	11.9	0.0002
Tooth adhesiveness	10.4[a]	7.5[c]	8.6[b]	65.7	<0.0001

Superscript letters indicate significant ($P < 0.05$) differences among treatment means in each row. Hardness was evaluated at first bite; all other sensory terms were evaluated during mastication where panelists were instructed to chew naturally.
[1]Number in parentheses indicates number of chews prior to evaluating textural attribute.

For the oral processing data, a repeated measures experimental design was used, where random effects were included for subject and subject-by-treatment interactions. The significance level was set at $\alpha = 0.05$ for all responses. No corrections were made to accommodate multiplicity. Correlations among rheological, sensory and oral processing parameters were evaluated using multivariate analysis in JMP 12.0. Pearson product-moment correlations indicate positive and negative correlations for positive and negative values, respectively, and significance levels are indicated at $P \leq 0.05$, $P \leq 0.01$, and $P \leq 0.001$.

Results and Discussion

Sensory

The goal of this study was to modify sensory texture with minimal changes to product formulation. Texture modifications were made using agar and gelatin, which replaced an equal weight of corn syrup. To facilitate presentation, caramels containing agar or gelatin are herein referred to as agar or gelatin caramels, respectively. The substitution of agar or gelatin for corn syrup did not significantly ($P > 0.05$) alter moisture or T_g, in agreement with previous reports for caramel formulated with similar biopolymer usage levels (Williams and others 2004). The 3 caramel samples averaged 9.8 ± 0.3% moisture with a glass transition (T_g) mid-point of −4.8 ± 1.2 °C (data not shown).

Sensory texture results are given in Table 3. Hardness, stickiness, and tooth adhesiveness were the most discriminating sensory terms. Agar caramels were perceived as hardest, followed by gelatin and control. The reverse order was true for stickiness and tooth adhesiveness. Cohesiveness was less discriminating among samples, in agreement with previous evaluations of cheese (Drake and others 1999) and caramel (Steiner and others 2003). Cohesiveness of a bolus is thought to be the contributing factor to initiating swallowing (Prinz and Lucas 1997; Agrawal and others 1998). Agar caramels were significantly less cohesive after 13 to 15 chews than gelatin and control, but no differences were seen among samples after 30 chews. Cohesiveness was relatively constant for agar and control when compared between the 2 extents of chewing, whereas gelatin cohesiveness was lower after 30 chews. This is likely due to gelatin melting at elevated oral temperatures, which has been shown to alter sensory perception of gels (Muñtoz and others 1986).

Sample cohesiveness did not differ significantly after 30 chews, which corresponded to ~40% to 60% of the total number of chews required to consume each sample. This lack of differentiation may be due to the inherent cohesive nature of caramels. The traditional oral processing paradigm considers foods that fracture into particles, with the goal being to reduce particle size and generate a cohesive bolus that can be safely swallowed (Prinz and Lucas 1995, 1997). However, when particle size reduction does not occur (for example, highly cohesive foods), other material transformations may play a larger role and need to be considered to interpret oral processing behavior (Brown and Braxton 2000). For caramel in particular, this includes phase transition of fat, dissolution of sugars, activity of salivary α-amylase, and softening of the mass.

Small amplitude oscillatory rheology

Rheological properties can be related to microstructural elements, which may then explain differences in macroscopic properties experienced during oral processing. The most common approach to evaluate texture using instrumental methods is to either *deform* (for example, compression, extension, bending, or torsion) or *punch* the sample and analyze various aspects of the force–deformation (or stress–strain) curve (Harker and others 2002; Barrangou and others 2006; Kim and others 2009; Çakir and others 2012c). However, caramels are problematic in that they show mainly ductile flow and do not fracture, thus limiting the types of mechanical attributes that can be used. Therefore, a combination of techniques was used to evaluate the mechanical fingerprint of caramels.

The frequency dependence of caramel samples under small amplitude oscillation is shown in Figure 1. Storage moduli (G') and loss moduli (G'') exhibited frequency-dependent behavior that increased with increasing frequency. As expected for a viscous, amorphous material, G'' was greater than G' for most of the frequency range until the values crossed at approximately 60 Hz (agar), 100 Hz (control), or 200 Hz (gelatin). The crossover is likely indicative of an entangled polymer network (Steffe 1996). This crossover range is higher than the 4 Hz crossover reported by Ahmed and others (2006), which can be attributed to differences in formulation and testing temperature. The phase angle (δ), representing the strain response that is in phase (0°) or out of phase (90°) with the stress input, indicated that all samples behaved as highly viscous fluids at frequencies <1 Hz (Figure 1).

Table 4–Rheological properties determined by creep recovery.

Rheological parameter	Control	Agar	Gelatin
J_0 (Pa^{-1})	2.96E-05[a]	3.06E-05[c]	2.74E-05[b]
J_{max} (Pa^{-1})	8.15E-04[a]	6.47E-04[c]	7.53E-04[b]
λ_{ret} (s)	34.2[a]	29.6[b]	32.8[a]
crp (%)	29.0[a]	47.7[b]	30.4[a]

Initial compliance (J_0), maximum compliance (J_{max}), retardation time (λ_{ret}) and recovery percentage (crp) of caramel formulations at 22 °C. Letters indicate significant differences ($P < 0.05$) among treatment means in each row.

Creep recovery

Properties determined by oscillatory testing showed that the caramel microstructure could be modeled as an entangled polymer network, but did not differentiate among formulations. Creep recovery testing was therefore used to provide additional rheological information measured over greater deformations and longer times (Table 4). Agar and gelatin caramels had significantly lower maximum compliance (J_{max}), indicating that they deformed less under a constant load. This is consistent with a firmer material that required more force (sensory hardness) to bite through. The agar caramel had a significantly shorter retardation time (λ_{ret}) and higher percent recovery (crp), both of which are indicative of greater elasticity. The creep recovery results coincide with some elements of the mechanical spectra. Lower J_{max} and higher crp for the agar treatment indicated a more elastic network that recovered after the strain, which coincided with a higher G' at very low frequency, and the lowest crossover frequency.

Pressure sensitive tack testing

Stickiness, tooth packing, and tooth adhesiveness are all related to the adhesion of the caramel to oral surfaces during mastication. Mechanical evaluations of these parameters fall under the field of pressure sensitive adhesion where adhesion is related to (1) the rheological properties of the adhesive and (2) surface energy differences between the adhesive and oral surfaces (Heddlesen and others 1993). Dahlquist (1989) reported a rheological criterion for tack, stating that adhesion does not occur when the adhesive material has a $G' > 100$ kPa. Furthermore, maximum adhesion is thought to occur for materials with G' from 50 to 100 kPa and a T_g between –10 and 10 °C (Foley and Chu 1986). All caramels fell within this maximum tack region over the frequency range 1 to 10 Hz. However, the frequency dependence of G' makes it difficult to examine relationships with sensory data. Rather than selecting G' at several frequencies, the inverse of J_{max} can be used to approximate a singular G' value. It should be noted that $1/J_{max}$ only represents an approximation of G' and is only being used for comparative purposes, as J_{max} reflects both elastic and viscous rigidity. Based on $1/J_{max}$ values, all 3 samples met the Dahlquist criterion for pressure-sensitive adhesion. Moreover, the agar caramel (1.5 kPa) was closer to the 100 kPa Dahlquist limit than gelatin (1.3 kPa) or control (1.2 kPa). This trend inversely correlates with

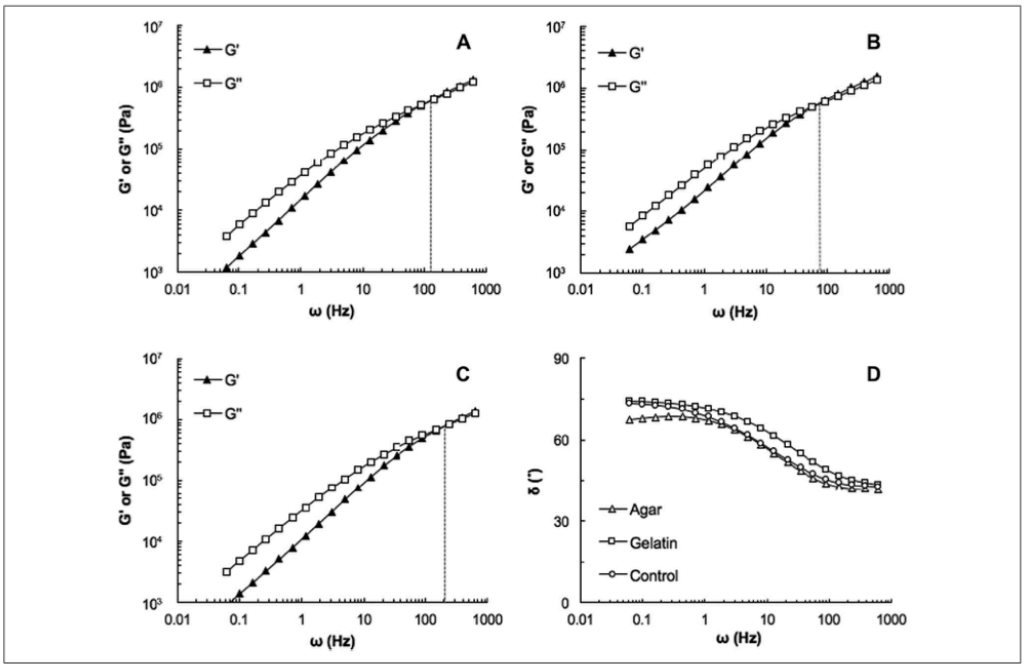

Figure 1–Mechanical spectra of caramels as determined via small amplitude oscillatory rheology. Storage modulus (G') and loss modulus (G'') of control (A), agar (B), and gelatin (C) caramels as determined by oscillatory rheology at 22 °C. Dashed line represents the crossover frequency of G' and G''. Phase angle (δ) of caramel formulations (D) were determined via small amplitude oscillatory rheology at 22 °C. Lines connecting data points are added as a visual aid.

sensory adhesiveness, where higher G' (approximated via $1/J_{max}$) is associated with lower adhesiveness.

Pressure sensitive adhesion is commonly measured by the application of a fixed surface area at a given force (compression) for a set amount of time (adherence) and then measuring the force required to separate the probe from the sample (decompression). In the context of oral processing, tack in the mouth could be related to the ability to make contact with oral surfaces during chewing. A constant normal force (20 N) was applied, which caused small differences in deformation, but produced a linear relationship between mechanical and sensory adhesion (Figure 2). It is important to note that these tests were performed without saliva and at temperatures below body temperature, which are recommended conditions for correlating with intra-oral adhesion due to the effects of saliva and temperature on surface energetics and rheology, respectively (Brenner and Nishinari 2014). However, without adjusting to intra-oral conditions, sensory adhesiveness—determined after the caramel was mixed with saliva and extensively processed—showed an excellent correlation with pressure sensitive tack.

Associations between sensory texture and mechanical properties

The first step in evaluating the relationship between food structure and texture is to examine correlations between mechanical parameters and sensory terms that have a logical link. Correlations among sensory terms and mechanical properties of caramels are shown in Table 5. Mechanical properties determined in creep recovery testing correlated with 5 of the 6 sensory terms, while properties measured in oscillatory testing showed no correlation. Oscillatory values at a frequency of 1 Hz were used for correlations as it most closely approximates chewing frequency (Prinz and Lucas 1995; Peyron and others 2002; Foster and others 2006; Koç and others 2014). Oscillatory testing is restricted to force, deformation, and time frames that keep the material within the linear viscoelastic region (Steffe 1996). This reflects probing the food structure without causing any changes that are not recovered within the timeframe of the test. In comparison, the applied stress (1 kPa) used in the creep recovery test was not restricted to the linear viscoelastic region.

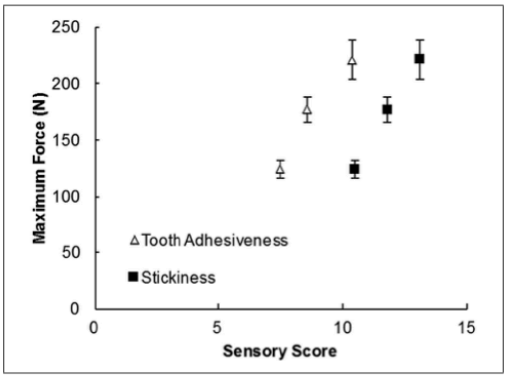

Figure 2–Relationship between sensory adhesion scores and pressure sensitive tack measurements. Comparison of sensory scores and pressure sensitive tack testing. Sensory stickiness and tooth adhesiveness were evaluated by the Spectrum method of descriptive anaylsis. Maximum force represents that maximum value on the force curve during decompression of a sample.

Sensory hardness correlated positively with crp, and negatively with J_{max} and λ_{ret}. This is logical as hardness is measuring the resistance to deform, and lower values for J_{max} reflect less deformation under a constant stress. High crp and low λ_{ret} indicate more elastic behavior. As seen in Table 4, only the agar caramel was significantly more elastic than the others, but the gelatin caramel trended toward more elasticity and contributed to the correlations. Brown and others (2003) reported similar associations for young cheese, where sensory firmness correlated positively with crp and negatively with J_{max}. Furthermore, the authors reported that rheological properties determined in creep recovery were more highly correlated with sensory texture than oscillatory testing.

Cohesiveness at 13 to 15 chews followed the opposite trends of hardness and correlated positively with J_{max} and λ_{ret}, and negatively with crp. It is not clear if this is simply coincidental with the change in material properties or if there is an underlying structural connection with maintaining a cohesive structure. Mechanical tack correlated strongly with stickiness and tooth adhesiveness, and negatively with hardness. This supports the notion that differences in rheological properties among caramels are determining differences in tack, as hypothesized by Dahlquist (1989). Nevertheless, there is no reason to rule out some contribution from surface energy.

Oral processing

Mechanical properties characterize elements of food structure and therefore potential elements of food texture. Oral processing can be considered the muscle activity, jaw movement, and tongue manipulations required to transform the food structure into a bolus and prepare for swallowing. Foods with different textural fingerprints—that is, differences in properties that comprise texture perception—have different oral processing patterns. This concept has been demonstrated for cheese (Çakir and others 2012a; Melito and others 2013), confections (Foster and others 2006), and biopolymer gels (Çakir and others 2012b; Hayakawa and others 2014). Mean values for oral processing parameters of each caramel formulation are presented in Table 6. Significant differences among samples were found for 8 of 17 mastication parameters, which are subcategorized as those associated with chew cycle timing, jaw movement and muscle activity. As a general trend, agar caramels required an increase in these parameters, which was in accordance with sensory texture properties. Gelatin caramels typically had values that fell between the agar and control caramels, although in this case they were not statistically separate from either group.

Oral processing of agar caramels required a greater number of chews, longer chewing cycles, and longer closing duration. These parameters are congruent with the increased hardness of agar samples. Similar results have previously been reported for confectionary products (Peyron and others 2002; Foster and others 2006; Grigoriadis and others 2014). Foster and others (2006) also reported slower chewing frequency for harder caramel samples; this trend was observed in the present study although not at the $\alpha = 0.05$ significance level. A possible explanation for the lack of statistical significance is the fact that we averaged chewing frequency over the entire oral processing sequence rather than discrete stages. Jaw movements are thought to be controlled by a central pattern generator, which coordinates movement of the jaw and muscle activity and modifies the chewing pattern in response

Table 5–Significant Pearson correlation coefficients between mechanical properties and sensory terms.

Rheological parameter	Hardness	Cohesiveness (13–15)[a]	Stickiness	Cohesiveness (30)[a]	Toothpacking	Tooth adhesiveness
J_{max}	–0.96***	0.83**	0.95***			0.90***
crp	0.86**	–0.85**	–0.86**	–0.65*		–0.80**
J_0						
λ_{ret}	–0.87***	0.86**	0.89***			0.82**
G' (1 Hz)						
G'' (1 Hz)						
δ (1 Hz)						
η^* (1 Hz)						
Tack force	–0.94***	0.71*	0.93***			0.94***

[a]Number indicates number of chews at which attribute was evaluated. Maximum compliance (J_{max}), percent recovery (crp), instantaneous compliance (J_0) and retardation time (λ_{ret}) were determined via creep recovery testing. Storage modulus (G'), loss modulus (G''), phase angle (δ) and complex viscosity (η^*) were determined via oscillatory testing at 1 Hz. Tack force was determined via pressure sensitive adhesion testing. Only significant correlation coefficients are presented.
Significance level is indicated at $^*P < 0.05$, $^{**}P < 0.01$, and $^{***}P < 0.001$.

Table 6–Oral processing parameters of caramel samples over the entire chewing sequence.

	Treatment				
Mastication parameter	Control	Agar	Gelatin	F ratio	*P*-value
Chew cycle timing parameters					
Number of chews**	32.6[b]	40.8[a]	34.7[b]	6.8	0.0072
Chewing frequency (s^{-1})	1.03	0.97	0.98		
Chewing cycle duration (s)*	0.84[b]	0.92[a]	0.87[ab]	5.5	0.0156
Opening duration (s)	0.38	0.41	0.39		
Closing duration (s)**	0.34[b]	0.38[a]	0.35[b]	10.3	0.0014
Power stroke duration (s)	0.13	0.12	0.12		
Jaw movement parameters					
Opening velocity (mm/s)	73.1	69.7	68.5		
Closing velocity (mm/s)	76.3	70.9	73.6		
Vertical movement (mm)	25.7	26.5	25.6		
Anterior posterior movement (mm)	9.0	8.9	8.8		
Medial lateral movement (mm)*	10.1[b]	10.8[a]	10.5[ab]	3.6	0.0499
Muscle activity parameters					
Temporalis activity per sequence[1]**	23.6[b]	33.8[a]	26.0[ab]	6.4	0.0093
Masseter activity per sequence[1]	19.8	27.1	21.7		
Total activity per sequence[1]**	43.4[b]	60.9[a]	47.7[ab]	6.9	0.0071
Temporalis activity per chew*	0.72[b]	0.83[a]	0.75[ab]	4.1	0.0362
Masseter activity per chew	0.61	0.66	0.63		
Total activity per chew*	1.33[b]	1.49[a]	1.38[ab]	3.8	0.0442

Superscript letters indicate significant differences among means within each row form results of analysis of variance and Tukey's separation test, with P-values <0.05* and <0.01**. Mean values were determined from 9 subjects chewing 2 samples of each treatment.
[1]Muscle activity per sequence indicates the total summed activity of all chews for a given sample.

to textural properties of the food during mastication (Yamada and others 2005). Thus, sample hardness has a greater influence on chewing frequency during the early stages of oral processing (Brown and others 1998; Lassauzay and others 2000; Foster and others 2006). Formulation also had no significant effect on power stroke duration. The power stroke represents the duration of occlusal contact of the teeth and is typically associating with structural breakdown of foods. Increased adhesiveness of caramels has been associated with longer power stroke duration (Çakir and others 2012a); however, the authors adjusted fat content to control adhesiveness, whereas we kept fat composition constant and varied adhesiveness only by the replacement of corn syrup with agar or gelatin.

Of the 3 oral processing subcategories, changes in jaw movement were least discernible among treatments. Only medial lateral movement was significantly affected by formulation, with a slight increase for gelatin caramels and a significant increase for agar caramels, both of which trended with increased hardness. It is important to note that formulation changes in caramels can affect hardness and adhesiveness, as we have shown with sensory data. Both attributes are not always reported, which may explain some of the discrepancies in the literature. For example, hardness in confectionary products has been associated with greater vertical and lateral jaw movement (Peyron and others 2002), or greater vertical movement with no change in lateral jaw movement (Foster and others 2006). Moreover, increased adhesiveness has been associated with greater jaw movement in all 3 directions (Çakir and others 2012a). These studies suggest that both adhesiveness and hardness influence jaw movement, making it difficult to identify the individual contributions of each attribute towards jaw movement in the present study.

Surface electrodes allowed for the quantification of muscle activity during mastication. Activity, in this sense, represents the integration of muscle activity over time; within each panelist session, these values were scaled against the largest value recorded. Means are represented as activity per sequence (that is, over the entire chewing process) and per individual chewing cycle. Chewing the agar caramel required more temporalis and total muscle activity per sequence and per chew than the control, with gelatin means falling between the agar and control samples. The trends were also observed with masseter activity, albeit just outside of the $\alpha = 0.05$ significance level. This is logical, as food texture influences muscle activity of the temporalis more than the masseter (Mioche and others 1999; Peyron and others 2002; Melito and

Table 7–Significant Pearson correlation coefficients between oral processing and sensory parameters.

Mastication parameter	Hardness	Cohesiveness (13–15)[a]	Stickiness	Cohesiveness (30)[a]	Toothpacking	Tooth adhesiveness
Chewing cycle duration	0.88**		–0.76*			
Opening duration	0.83*		–0.81*			
Closing duration	0.76*		–0.84*			
Power stroke duration					0.86*	
Opening velocity						
Closing velocity	–0.82*		0.89**			0.81*
Vertical movement						
Maximum opening time	0.94**		–0.76*			
Anterior posterior movement					0.79*	
Medial lateral movement	0.87*					
Temporalis activity per sequence						
Masseter activity per sequence						
Temporalis activity per chew	0.79*		–0.77*			
Masseter activity per chew						
Number of chews	0.78*			–0.79*		
Chewing frequency						

[a]Number indicates number of chews at which attribute was evaluated. Only significant correlation coefficients are presented. Significance level is indicated at $^{*}P < 0.05$, $^{**}P < 0.01$, and $^{***}P < 0.001$.

others 2013). Increased muscle activity for agar caramels was concomitant with increased sensory hardness, which has been reported for a variety of foods (Jack and others 1993; Mathevon and others 1995; Mioche and others 1999; Foster and others 2006; Çakir and others 2012b). Likewise, higher adhesiveness of caramels at a constant hardness requires more muscle activity (Çakir and others 2012a). In these studies, the effect of 1 attribute was evaluated by holding other attributes constant. Conversely, in this investigation hardness and adhesiveness were inversely related, with the agar caramel being the hardest but least adhesive. It would appear that the net increase in muscle activity necessary to overcome hardness predominated the net decrease in muscle activity associated with reduced adhesiveness. It is intuitive that muscle activity is being used to manipulate caramels in order to overcome hardness and adhesiveness, so those sensory texture effects are convoluted in the muscle activity measurements.

The caramel formulations in this study were not modified to a great degree; therefore, the differences in oral processing measurements were subtle. However, compared to similar studies on the oral processing of biopolymer gels and cheese (Çakir and others 2012a; Koç and others 2014; Melito and others 2013), caramels have a markedly different oral process. This could be due to differences in adhesion and/or hardness. Most notably, the magnitude and velocity of jaw movement are greater, while chewing frequency is slower. In essence, the jaw has to travel a greater linear distance to overcome a greater degree of hardness and adhesiveness.

Relationships among oral processing parameters and sensory terms

As with mechanical properties, hardness was the primary sensory term that correlated with mastication parameters (Table 7). This is logical as hardness is associated with the jaw movement and muscle activity to accomplish the first chew. Hardness correlated with chewing duration, opening time, medial lateral movement, temporalis activity per chew and total number of chews. Similar correlations between hardness and muscle activity, chewing frequency and chewing duration have been reported for mixed biopolymer gels (Çakir and others 2012b).

Since adhesiveness was inversely related to hardness, it tended to correlate with the same mechanical (Table 5) and oral processing (Table 7) parameters as hardness but with a negative correlation. This included chewing duration, opening time and temporalis activity per chew. These correlations do not agree with the results of Çakir and others (2012b), who reported that adhesiveness of biopolymer gels only correlated with chewing frequency. In this study, hardness and adhesiveness were not independently controlled. Compared to the aforementioned correlations in the literature, (that is, our hardness data agrees with literature whereas adhesiveness does not), it appears that the effects of hardness outweigh those of adhesiveness during oral processing, although that remains to be explicitly tested.

The one parameter that did not correlate with any mechanical property—tooth packing—correlated with power stroke duration and anterior posterior movement. This suggests that jaw movements are adjusted to deal with tooth packing. There were minimal differences in tooth packing among treatments (Table 3), so a greater range in this texture parameter would need to be investigated.

Conclusions

Minor changes in the polymer composition of caramels produced significant changes in sensory texture. Hardness was associated with material properties based on force–deformation (J_{max}) relationships and the muscle activity needed to close the jaw (temporalis activity per chew). Likewise, the magnitude of pressure sensitive adhesion, although determined in the absence of saliva, was linked with adhesion to oral surfaces (stickiness) and force required to remove caramels from tooth surfaces (tooth adhesiveness). Jaw movements previously associated with increased hardness or adhesiveness were not observed, most likely due to hardness and adhesiveness changing inversely among the samples. This would suggest that jaw movement patterns need to be analyzed relative to the oral processing requirements and associated texture terms of the food material. In this case, changes in movement associated with decreases in hardness were coupled with adjusting to increased adhesiveness. It is recommended that a combination of material properties and oral processing parameters be used to gain a more complete understanding of factors determining sensory texture perception.

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National Inst. of Food and Agriculture. Support from the North Carolina Agricultural Research Service is gratefully acknowledged. The use of trade names in this publication does not imply endorsement by the North Carolina Agricultural Research Service of the products named or criticism of similar ones not mentioned. The authors are very grateful for supplies donated by Fonterra, Vyse and TIC Gums. Additionally, the authors acknowledge assistance from Chris Vinyard with oral processing, MaryAnne Drake and Michele Yates for sensory analysis, Emily Griffith for support with statistical design, and Chris Pernell for rheological analysis.

Contributions

TBW, PJL, and EAF designed the study and interpreted the results. TBW collected test data and drafted the manuscript. PJL and EAF edited and revised the manuscript.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

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Supporting Information

Additional supporting information may be found in the online version of this article at publisher's website:

Table S1. Definitions of oral processing terms.

CHAPTER 7. SURFACE ENERGY AND VISCOELASTICITY INFLUENCE CARAMEL ADHESIVENESS

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RESEARCH ARTICLE

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Surface energy and viscoelasticity influence caramel adhesiveness

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Abstract

Adhesion is an important textural attribute that directs consumer eating patterns and behaviors and can be a negative attribute during food processing. The objectives of this study were to modify caramel formulation and compare adhesion to different materials to quantify the influence of surface energetics and viscoelasticity on caramel adhesiveness. Mechanical adhesion was viewed in the context of pressure sensitive tack theory, where adhesion is controlled by viscoelasticity of the adhesive material and the surface energy relationship of material and probe. Caramel samples varied in total amount of fat and protein, and mechanical adhesion was measured using a series of materials with total surface energies of 39.7–53.2 mJ/m^2. Adhesiveness decreased as fat and protein content increased, with a significant effect of total surface energy. Viscoelasticity was modeled using creep recovery data fit to a four-element Burger mechanistic model. Burger model parameters representing retarded elasticity correlated strongly with adhesiveness. The results suggest two zones of adhesion based on formulation, one driven by both surface energy relationships—most notably dispersive and total surface energy—and viscoelasticity, and the other driven solely by viscoelasticity.

Practical applications

Relationships between mechanical properties and adhesion have been explored but are still not well understood, and could aid in the design of food products with a controlled level of adhesion. The results of this study indicate the importance of considering material surface energy when measuring mechanical adhesion or texture profile analysis. Understanding the relationships between viscoelastic behavior and adhesion can be used to make inferences on perceived texture.

KEYWORDS
adhesion, confections, creep test, surface energy, texture

1 | INTRODUCTION

Adhesiveness is an important physical property to consider when designing foods. Alina Szczesniak first defined sensory adhesiveness as one of the five primary mechanical characteristics of texture (Szczesniak, 1963). Likewise, a mechanical measure of adhesiveness is part of the original mechanical texture profile analysis method using a General Foods Texturometer (Friedman, Whitney, & Szczesniak, 1963) and Malcolm Bourne's adaptation of the method to a universal uniaxial testing machine (Bourne, 1978). Adhesiveness can be considered a negative attribute of a food, where adhering to processing surfaces can be troublesome (Saunders, Hamann, & Lineback, 1992), or during consumption, where adhesiveness influences eating rate and eating difficulty (Çakır et al., 2012; Hayakawa et al., 2014), most notably with the elderly (Peleg, 1993). However, adhesiveness can also be a positive attribute and even expected for certain types of foods, including cheese or confections such as caramel. It may also be an important attribute in determining if a food is ready to be swallowed (Loret et al., 2011). Therefore, understanding the mechanisms determining adhesion and how to control it are important to food processing and addressing the needs of consumers.

In a general sense, adhesiveness describes the propensity of two surfaces—often referred to as adhesive and adherend—to adhere to each other. Adhesiveness of many foods can be explained using

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pressure sensitive adhesive theory, meaning the materials are viscoelastic and exhibit strong cohesiveness, and adhesion no longer occurs when the adhesive storage modulus exceeds 10^5 Pa (Dahlquist, 1989). The magnitude of pressure sensitive adhesion is governed by material properties (e.g., rheology, glass transition temperature, ingredient hydrophobicity, and surface chemistry) and external conditions such as applied pressure, contact time, temperature, contact surface area, and presence of lubricant (Adhikari, Howes, Bhandari, & Truong, 2001; Bhandari & Howes, 2005; Foley & Chu, 1986; Saunders et al., 1992). Of these factors, rheology and interfacial surface energies are considered the predominant contributors to pressure sensitive adhesion (Zosel, 1989).

It has long been the goal of the food industry to have instrumental tests to predict adhesiveness for processing and sensory considerations. These tests are often empirical in nature and commonly include compression testing or texture profile analysis. They are generally approached within the confines of an adhesive in contact with an adherend under a given set of conditions that vary across sample types (Adhikari et al., 2001; Brenner & Nishinari, 2014; Dunnewind, Janssen, Van Vliet, & Weenen, 2004; Kilcast & Roberts, 1998; Kim et al., 2009; Michalski, Desobry, & Hardy, 1997). These tests appear to be most applicable to food processing conditions where food materials are in contact with various surfaces during manufacturing. In contrast, instrumental predictions of sensory adhesiveness (defined as the degree to which a product sticks to teeth and oral surfaces after initial contact, several chews, or throughout mastication) have been less successful. This is likely because there is no one single adhesiveness value vis-à-vis oral processing, where material transformations modify the sensation from first bite through swallowing (Brenner & Nishinari, 2014; Wagoner, Luck, & Foegeding, 2016). As a result, correlations between mechanical and sensory measures of adhesiveness tend to be product specific and there is currently no single theory to predict adhesion across multiple types of foods (Adhikari et al., 2001; Bhandari & Howes, 2005; Saunders et al., 1992).

It has been previously shown that adhesiveness is a dominant textural attribute of caramels and instrumental adhesion correlates strongly with sensory adhesiveness (Kilcast & Roberts, 1998; Steiner, Foegeding, & Drake, 2003; Wagoner et al., 2016). As a result, caramel was selected as a model system for this study. Caramel is an emulsion of a low water, high solids continuous phase (sucrose, lactose, small saccharides, polymers, casein, and whey proteins) and a semi-crystalline fat dispersed phase, with denatured milk proteins and lecithin acting at the oil-water interface (Heathcock, 1985). Amorphous caramels have little to no sugars in a crystalline state and typically exhibit fluid-like viscoelastic behavior (Hartel, Ergun, & Vogel, 2011). The amount and type of fat is important for flavor as well as reducing adhesiveness (Mendenhall & Hartel, 2016). Altering the types and ratios of the ingredients modifies bulk textural attributes and is used to generate different types of caramels (Steiner et al., 2003; Wagoner et al., 2016).

This study seeks to build on previous research showing the effects of caramel formulation on mechanical properties, sensory attributes, rheology, and oral processing (Mendenhall & Hartel, 2016; Steiner et al., 2003; Wagoner et al., 2016). Creep recovery tests were used as a rheological tool because they are useful for explaining time-dependent viscoelastic behavior. We hypothesize that caramel adhesiveness is a dynamic property influenced by (a) the underlying mesostructure that is controlled by formulation and reflected in rheological properties, and (b) the interfacial energy between caramel and another contacting surface. To our knowledge, these relationships have not been fully explored. Therefore, the objectives of this study were to modify surface type and caramel formulation to quantify the influence of surface energetics and viscoelasticity on caramel adhesiveness.

2 | MATERIALS AND METHODS

2.1 | Sample formulation

Anhydrous butter fat containing > 99.6% fat was provided by Land O'Lakes (Arden Hills, MN). Corn syrup with a dextrose equivalence of 42 and soy lecithin were provided by Cargill Inc. (Minneapolis, MN). Granulated cane sugar (Domino, Domino Foods Inc., Iselin, NJ) and sweetened condensed skim milk (SCSM; Eagle Brand, J.M. Smucker Co., Orrville, OH) were purchased at a local grocery.

Caramel formulations varied by total amount of protein and lipid sources (denoted as PL%) by adjusting the amounts of SCSM and butter fat (formulations in Table 1). A constant 2:1 mass ratio of SCSM to butter fat was maintained to ensure that the ratio of protein to fat was held constant. All other ingredients were scaled proportionally. Caramels were prepared in 600 g batches according to the method described by Steiner et al. with several modifications (Steiner et al., 2003). In short, milk fat was melted in a stainless-steel pan (All-Clad, Metal Crafters Inc., Cannonsburg, PA) on a 180C hot plate. The remaining ingredients were added and mixed with an overhead mixer (Heidolph Instruments, Schwabach, Germany) fitted with a custom stainless steel paddle (135 mm diam.) at 320 rpm for 20 min to emulsify the fat before increasing the heat to 375C until the mixture reached 118C. Caramel was removed from the heat and mixed by hand for 10 s before pouring into cylindrical molds (19 mm height, 19 mm diameter) lined with acetate film to prevent formation of a surface skin. Each batch was cooled for 60 min and packaged for storage to minimize

TABLE 1 Caramel formulations by total PL (% W/W)

	10%	20%	30%	40%
Water	19.1	16.9	14.8	12.7
Sucrose	38.9	34.6	30.3	26.0
Corn syrup	31.8	28.3	24.7	21.2
SCSM	6.7	13.3	20.0	26.7
Butter fat	3.3	6.7	10.0	13.3
Soy lecithin	0.2	0.2	0.2	0.2
Total	100	100	100	100

Notes. SCSM is sweetened condensed skim milk. PL represents total amount of protein source (SCSM) and lipid source (butter fat).

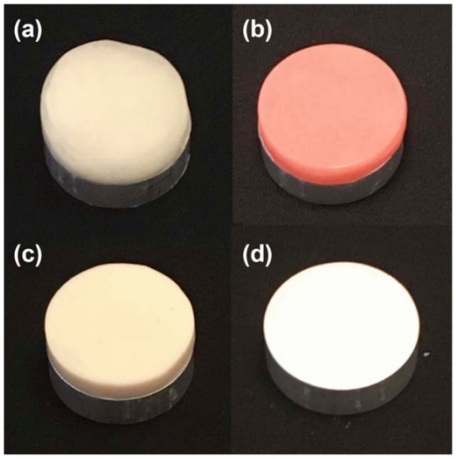

FIGURE 1 Surfaces used for pressure sensitive adhesion tests. Restorative composite (a), denture acrylic resin (b), temporary crown acrylic (c), and brushed aluminum (d) surfaces (35 mm diameter)

moisture loss. Caramels were stored at room temperature (herein 22 ± 1C) for at least 12 hr and no more than 48 hr prior to analysis.

2.2 | Surfaces (adherends)

Four surfaces were used for pressure sensitive adhesion evaluation (Figure 1), including brushed aluminum and three dental analogs: denture acrylic resin (Lucitone 199, Dentsply International, York, PA), temporary crown acrylic (Integrity, Dentsply International), and composite restorative material (PermaFlo, UltraDent Products, Inc., South Jordan, UT). All surfaces were manufactured into cylinders (35 mm diameter) and smoothed using 180-grit sandpaper (3M, St. Paul, MN) to reduce surface roughness as a confounding variable.

2.3 | Surface energy

The sessile drop method was used to determine the contact angle of droplets on the surface of each material (Owens & Wendt, 1969). Water and methylene iodide were used as probe liquids. An 8 μL drop of each probe liquid was deposited onto the material surfaces and images were obtained using a contact angle goniometer (Ramé-Hart Instrument Co., Succasunna, NJ). Contact angle was determined using DROPimage software (Ramé-Hart Instrument Co.). Mean contact angle values were the average of twelve independent measurements taken on two separate days. Material surface energy was calculated using Equation 1, where θ is the contact angle, σ is the surface energy/surface tension (units), L represents liquid, S represents the solid material, P represents polar contribution to total surface energy, and D represents dispersive contribution to total surface energy.

$$\frac{\sigma_L(\cos\theta + 1)}{2\sqrt{\sigma_L^D}} = \frac{\sqrt{\sigma_S^P}\sqrt{\sigma_L^P}}{\sqrt{\sigma_L^D}} + \sqrt{\sigma_S^D} \quad (1)$$

The contribution of dispersive forces was determined using Equation 2 and the assumption that methylene iodide has no polar interactions with the surface. The difference in dispersion energy and total surface energy is reported as the contribution of polar interactions.

$$\sigma_L^D = \frac{(\sigma_L[\cos\theta + 1])^2}{72} \quad (2)$$

2.4 | Compressive hardness and pressure sensitive adhesion

Mechanical aspects of texture were evaluated on a TA XT Plus texture analyzer (Stable Micro Systems, Godalming, UK). Hardness was evaluated by compressing cylindrical samples (19 mm height, 19 mm diameter) using the aforementioned aluminum surface (35 mm diameter). Before each individual measurement, the top layer of each sample was removed using a knife to ensure a flat surface and to eliminate any skin. Hardness was the maximum force (N) obtained during compression to 65% of original height at 5 mm/s. This allowed the sample to deform without exceeding dimensions of the upper probe. Pressure sensitive adhesion was measured according to the method described by Wagoner et al. (2016) with several modifications. To account for differences in hardness, the samples were compressed to a constant normal force of 200 N. The probe was lowered at a rate of 5 mm/s, compressed to 200 N, held for 10 s, and withdrawn at 10 mm/s. Adhesiveness represents the maximum force (N) required to remove the probe from the sample. Hardness and adhesiveness measurements were both repeated in triplicate, and the arithmetic mean of four samples was used for each independent replicate.

2.5 | Creep recovery

Creep recovery tests were conducted on a controlled stress rheometer (Anton Paar MCR 302, Graz, Austria) equipped with 25 mm diameter parallel plates. All tests were conducted within the linear viscoelastic region for samples (based on preliminary analysis). Prior to each test, caramels were cut into discs (16 mm diameter, 5 mm height) and compressed to a 2 mm gap between plates. Samples were equilibrated to 0.1 N normal force for 120 s. During the creep portion of the test, a constant 1 kPa shear stress was applied for 60 s; the sample was then allowed to recover for 360 s. Data were fit to a four element Burger's model (Equation 3), where instantaneous compliance (J_0) represents the first compliance value after the stress was applied, J_1 represents the retarded compliance, η_N represents Newtonian viscosity of the free dashpot, and retardation time (λ_{ret}) is the time required to reach 63.2% of maximum compliance (Steffe, 1996). Percent recovery (crp) is the percentage of compliance recovered during relaxation as determined using Equation 4, where J_{max} represents the maximum compliance during the creep portion of the test, and J_r represents the minimum compliance value during relaxation.

$$J_{(t)} = J_0 + J_1\left(1 - e^{\left(\frac{-t}{\lambda_{ret}}\right)}\right) + \frac{t}{\eta_N} \quad (3)$$

$$crp = \frac{J_{max} - J_r}{J_{max}} \times 100 \quad (4)$$

2.6 | Statistical analysis

Statistical analysis was performed using JMP Pro software version 13.0 (SAS, Cary, NC). Treatment effects were determined using analysis of variance (ANOVA) with Tukey's HSD (experiment–wise error set at $\alpha = 0.05$ for each response) as a post hoc multiple comparisons test to separate means where necessary. Pearson correlation coefficients were determined using multivariate analysis with pairwise comparisons, with significance indicated at $^{*}p < .05$ and $^{**}p < .01$.

3 | RESULTS

3.1 | Surface energy of adhesive surfaces

Pressure sensitive adhesiveness is a product of adhesive rheology and surface energy, which indicates how a viscoelastic fluid wets the surface of a material (Michalski et al., 1997). Four surfaces were selected based on similarities to processing surfaces (aluminum) or use as artificial dental surfaces (crown acrylic, denture acrylic, and restorative composite), and to cover a range of dispersive, polar, and total surface energies. Dispersive contributions indicate weak van der Waals type interactions, where all materials have at least some portion of dispersive surface energy; polar contributions represent hydrogen bonding and dipole–dipole interactions (Kilcast & Roberts, 1998).

Total surface energy of all surfaces ranged from 39.7 to 53.2 mJ/m^2 (Table 2). Both acrylics had surface energies greater than 50 mJ/m^2, although they differed slightly in polar surface energy. Aluminum and composite surfaces both had total surface energies below 40 mJ/m^2 and low polar surface energies. Adhesiveness is thermodynamically favorable when polar surface energies are very similar (Wu, 1973) or at high adherend surface energy and low adhesive surface energy (Owens & Wendt, 1969).

3.2 | Caramel hardness

Caramel formulations varied in the total amount of protein contributed from the SCSM and butter fat, herein noted as PL% for brevity. The ratio of these ingredients was kept at a constant 2:1 mass ratio to ensure that the ratio of protein to fat was held constant; the ratio of all other ingredients was scaled proportionally. Caramel hardness was evaluated as the maximum force required for a constant compressive distance (Figure 2). Hardness decreased with increasing PL up to 30%, with no difference between 30 and 40% PL. This trend aligns with a previously reported inverse relationship between hardness and fat content (Mendenhall & Hartel, 2016).

TABLE 2 Dispersive, polar, and total surface energy of probe materials used in adhesion testing (mJ/m^2)

	Aluminum	Crown acrylic	Composite	Denture acrylic
Dispersive	37.5 ± 0.9	47.9 ± 0.7	37.1 ± 1.0	47.5 ± 0.4
Polar	2.2 ± 0.3	4.1 ± 0.4	0.7 ± 0.3	5.7 ± 0.3
Total	39.7 ± 0.8	51.9 ± 0.8	37.8 ± 0.8	53.2 ± 0.5

Note. Data are presented as means ± standard deviation from twelve measurements.

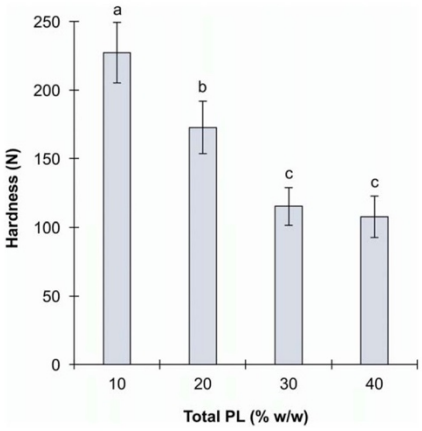

FIGURE 2 Mechanical hardness of caramel formulations based on total PL (% W/W). Determined by compressing samples (19 mm height, 19 mm diameter) using an aluminum probe (35 mm diameter). Letters indicate significant differences in means as determined by ANOVA followed by Tukey's HSD post hoc multiple comparisons

3.3 | Caramel adhesiveness

Because hardness was not constant across samples, adhesiveness was measured at a constant compressive force to compensate. It is also important to note that all samples exhibited adhesive failure; that is, the samples had enough cohesive strength to remain as a single piece and separate cleanly from the probe. Separation of the upper probe from samples occurred rapidly, therefore, maximum adhesive force was used instead of tack energy or the total area under the debonding curve.

Increasing PL up to 30% resulted in lower adhesiveness for all surfaces (Figure 3). Similar trends have previously been reported for caramel, where higher amounts of palm kernel oil and SCSM were associated with reduced sensory and mechanical adhesiveness (Steiner et al., 2003). Increasing PL from 30 to 40% was associated with a slight increase in adhesiveness for all surfaces.

At 10 and 20% PL, adhesiveness was significantly greater with the acrylics as compared to the aluminum and composite surfaces. However, this trend no longer held true above 30% PL, where adhesiveness was highest with the crown acrylic and lowest with the denture acrylic. This divergence in trends of low PL (10 and 20%) compared to high PL

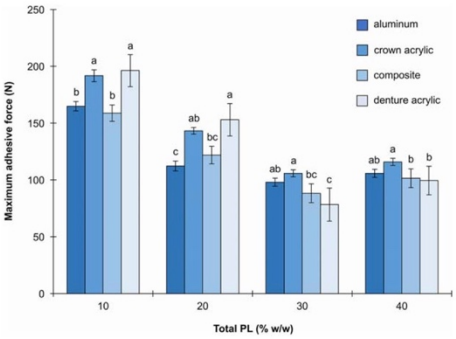

FIGURE 3 Caramel adhesiveness based on total PL (% W/W) and probe surface material type. Letters indicate significant differences in means within each PL group as determined by ANOVA followed by Tukey's HSD post hoc multiple comparisons

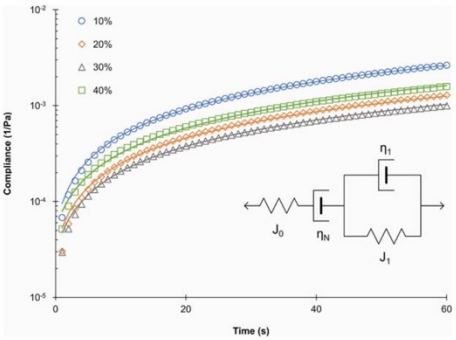

FIGURE 4 Creep curves fit to Burger's four-element model by total PL (% W/W). Symbols represent creep data and solid lines represent four-element Burger's model fit to data. Inset schematic represents four-element Burger's model, where J_0 denotes instantaneous elastic compliance, J_1 denotes retarded compliance, η_N denotes long-term Newtonian viscosity, and η_1 denotes the retarded viscous element

(30 and 40%) was evident in correlations among surface energies and adhesion (Table 3). Surface energy correlated strongly with adhesiveness at 10% PL (dispersive, polar, and total: $R^2 > 0.97$) and 20% PL (dispersive and total: $R^2 > 0.94$), whereas no significant correlations were observed at 30 and 40% PL. Thus, it appears that all aspects of surface energy (polar, dispersive, and total) can be useful in predicting caramel adhesiveness but only below a certain PL threshold.

3.4 | Creep recovery

Surface energy explained at least a portion of differences in adhesion within each formulation, but changes in rheological properties with PL will also influence adhesiveness. To gain an understanding of how rheological properties influence adhesiveness, creep recovery was used. Creep recovery probes the time dependent viscoelastic behavior of samples and has previously been shown to measure properties that distinguish differences among caramel formulations (Wagoner et al., 2016). It can be used to probe higher strains and longer times, which are more representative of oral processing conditions (Foegeding, Brown, Drake, & Daubert, 2003). This is an important consideration for relating sensory and instrumental measures of adhesiveness (Brenner & Nishinari, 2014).

Results from creep recovery are given in Figure 4. The 1 kPa shear stress applied during the creep portion was determined to fall within the linear viscoelastic region. Creep curves were separated into discrete attributes (Table 4) to describe the viscoelastic response over time (Subramanian, Muthukumarappan, & Gunasekaran, 2003). These relate to the components of the four-element Burger's model: instantaneous elasticity, retarded elasticity, and viscous flow (Steffe, 1996).

In polymer systems, J_0 represents an instantaneous elastic compliance related to an undisturbed polymer network (Ma, Drake, Barbosa-Cánovas, & Swanson, 1997; Subramanian et al., 2003). J_0 decreased as PL increased up to 30% and then increased at 40%. A higher J_0 is indicative of instantaneous elastic behavior and polymer rearrangement during stress, meaning that more deformation is needed prior to flow (Jiménez-Avalos, Ramos-Ramírez, & Salazar-Montoya, 2005). This could be indicating a network; however, it does not scale with increasing amount of protein.

Sample retarded elasticity would be related to J_1 and λ_{ret} per the Burger's model, in addition to J_{max}. Maximum compliance, J_{max}, is a ratio of strain to stress and can be considered the inverse of firmness.

TABLE 3 Pearson correlation coefficients among surface energy and adhesiveness by total PL (% W/W)

	10%	20%	30%	40%
Dispersive	0.987**	0.941*	0.014	0.354
Polar	0.966*	0.881	0.254	0.068
Total	0.999**	0.942*	0.080	0.283

Note. Statistical significance indicated at $p < .01$ = ** and $p < .05$ = *.

TABLE 4 Creep recovery parameters by total PL percentage (% W/W)

	10%	20%	30%	40%
η_N (Pa·s × 10^4)	2.35d	5.09b	6.71a	4.16c
J_{max} ($Pa^{-1} \times 10^{-5}$)	2.64a	1.29c	1.00d	1.58b
J_1 ($Pa^{-1} \times 10^{-5}$)	2.96b	6.84a	7.62a	8.45a
J_0 ($Pa^{-1} \times 10^{-5}$)	5.43a	3.05b	2.96b	5.19a
J_0* (%)	2.05d	2.37c	2.97b	3.29a
J_1* (%)	1.12c	5.32b	7.64a	5.36b
λ_{ret} (s)	37.1a	36.0b	35.6b	35.7b
crp (%)	7.8d	11.6c	20.9a	18.5b

Note. Letters indicate significant differences in means within each row as determined by one way ANOVA and Tukey's HSD post hoc multiple comparisons.

Caramels made with 10% PL had the greatest J_{max}, which decreased with PL up to 30% and then increased at 40%. Retardation time (λ_{ret}) was longest for the 10% PL sample, whereas no significant differences were observed among 20, 30, and 40% PL. This indicated that it took longer for the applied stress to deform the network at 10% PL as the material was deforming more slowly (Steffe, 1996). Network elasticity and λ_{ret} are inversely related (Ojijo et al., 2004).

Mechanical aspects of the system can also be determined by expressing J_0 and J_1 as percentages of maximum compliance (Equations 5 and 6), where J_0^* indicates elastic contributions and J_1^* indicates viscoelastic contributions to compliance (Dolz, Hernandez, & Delegido, 2008; Olivares, Zorrilla, & Rubiolo, 2009). All samples had relatively low values for both, confirming primarily viscous behavior in the system. Only at 10% PL was J_0^* greater than J_1^*; all other samples had J_0^* less than J_1^*, suggesting slightly more elastic behavior at 10% PL.

$$J_0^* = \frac{J_0}{J_{max}} \times 100 \quad (5)$$

$$J_1^* = \frac{J_1}{J_{max}} \times 100 \quad (6)$$

Long-term viscous flow and irrecoverable deformation would be described by η_N (i.e., Newtonian viscosity of the free dashpot) and creep (crp), where purely elastic Hookean solids and Newtonian fluids exhibit 100 and 0% recovery, respectively. The η_N increased with PL up to 30% then decreased at 40% PL. All samples exhibited primarily viscous behavior as shown by low recovery of energy (crp), which has previously been reported for amorphous caramels at low frequencies during small amplitude oscillatory testing (Ahmed, Ramaswamy, & Pandey, 2006; Wagoner et al., 2016; Weir, Bromley, Lips, & Poon, 2016).

3.5 | Correlations among mechanical properties and creep recovery parameters

Mechanical adhesiveness correlated strongly with λ_{ret} and negatively with J_1, J_1^*, and crp (Table 5). These results are similar to caramels modified with hydrocolloids equivalent to 30% PL in this study (Wagoner et al., 2016). Adhesiveness also correlated strongly with J_{max}, which is intuitive in the sense that a more compliant material would better wet microscopic fissures on a surface; wetting is a key component to determining how well a liquid will adhere to a solid surface (Michalski et al., 1997). Similar correlations of J_{max} and J_0—but not λ_{ret}—with tack energy have been reported for mozzarella cheese (Childs, Daubert, Stefanski, & Foegeding, 2007). The slight differences could be attributed to the protein network, where cheese has a higher protein concentration and greater degree of elasticity than caramel. Caramel hardness and adhesiveness strongly correlated with many of the same rheological parameters. This comes as no surprise as hardness and adhesiveness are themselves highly correlated, making it difficult to discern individual rheological contributions.

Within rheological parameters, λ_{ret} correlated negatively with η_N, J_1, and crp, which would be expected because these are related aspects of time-dependent viscoelasticity (Olivares et al., 2009). The negative correlation between η_N and J_0 is also intuitive in that they describe different aspects of the Burger model: J_0 as an instantaneous elastic response and η_N for long term viscous flow. Where J_0 did not correlate with many parameters, the transformation from discrete values to compliance as a percentage of maximum compliance (i.e., J_0^*) did correlate with other attributes. However, for caramel samples where J_0^* and J_1^* are less than 10%, it may be more telling to examine the relationship between J_0^* and J_1^* rather than the individual values (Olivares et al., 2009).

3.6 | Discussion

The results of this study indicate that even across a narrow range of surface energy (39.7–53.2 mJ/m^2), accounting for material surface energy and time-dependent viscoelasticity is important for understanding adhesion. For the caramel formulations in this study, increasing PL corresponded to additional fat and protein at the expense of sugars and polysaccharides. The decrease in adhesion with increasing PL comes as no surprise based on previous reports of reduced adhesiveness with increasing fat content (Steiner et al., 2003); this study builds

TABLE 5 Pearson correlation coefficients among adhesiveness, hardness, and creep recovery parameters

	Hardness	Adhesiveness	η_N	J_1	J_0	crp	λ_{ret}	J_1^*	J_0^*	J_{max}
Hardness	1.00									
Adhesiveness	0.81**	1.00								
η_N			1.00							
J_1	−0.75*	−0.76*		1.00						
J_0			−0.88**		1.00					
crp	−0.90**	−0.83**	0.76*	0.76*		1.00				
λ_{ret}	0.86**	0.78*	−0.83**	−0.91**		−0.86**	1.00			
J_1^*	−0.75*	−0.75*	0.92**	0.87**		0.84**	−0.95**	1.00		
J_0^*	−0.91**	−0.81**		0.83**		0.90**	−0.81**		1.00	
J_{max}		0.95**			0.82*	−0.77*	0.94**			1.00

Note. Omitted values were not statistically significant at $\alpha = 0.05$. Statistical significance indicated at $p < .001$ = ** and $p < .05$ = *.

on this knowledge to further characterize the mechanisms that control adhesion. The data suggest two zones of adhesiveness in caramels based on ratio of PL to carbohydrates, and the controlling mechanisms for adhesion depend on the zone. In the first zone, corresponding to 10 and 20% PL, adhesiveness was controlled by both viscoelastic behavior and surface energetics. In the second zone, adhesion was primarily driven by viscoelasticity. Similar two-zone mechanisms of adhesion have previously been described for cheese (Childs et al., 2007). With pressure sensitive adhesives, when the surface energy of an adherend is less than the adhesive, wetting is a function of viscoelasticity and surface energetics; when adherend surface energy is greater, it is primarily a viscoelastically driven phenomenon (Zisman, 1963).

Adhesion is thought to be maximized if an adhesive material can flow and fully wet a surface, where wetting is favored by low adhesive surface energy and high adherend surface energy (Michalski et al., 1997; Owens & Wendt, 1969). In this study, further information could be gleaned if the surface energy of the caramel formulations was known. However, this could not be experimentally determined due to absorption of the probe liquids on the caramel surface. Based on the two-zone mechanism of adhesion, we hypothesize that the surface energy of 10 and 20% PL caramels was greater than that of the probes. Adhesiveness would then decrease as the surface energy of the caramel approaches that of the probes (Zosel, 1989). Conversely, the surface energy of 30 and 40% PL caramels was near or below that of the probes, where adhesiveness would be primarily controlled by viscoelasticity independent of surface energy (Zisman, 1963).

Per the two-zone model of adhesion, rheology would be a contributor to adhesion across both zones, with more viscous behavior and deformability resulting in greater maximum tack energy (Gay, 2002; Heddleson, Hamann, & Lineback, 1993). However, small strain oscillatory parameters G', G'', δ, and η^* measured at a frequency similar to chewing rate (1 Hz), did not correlate with sensory adhesion in caramels, while creep recovery parameters were highly correlated (Wagoner et al., 2016). This may be related to sensory adhesion (i.e., stickiness and tooth adhesiveness) being evaluated during the chewing process and, therefore, over a longer time frame (Wagoner et al., 2016). Data were, therefore, fit to a four element Burger's model to evaluate various elements of viscoelasticity.

A comparison can be made between creep and pressure sensitive adhesion tests assuming the samples respond in similar ways, even though stress and strain values are different. First, a stress is applied as instantaneous as possible in creep testing and over the course of several seconds in shear. This makes the instantaneous strain (J_0) portion of creep very different than an adhesion test where strain is applied over time until a set stress is achieved, and may explain the lack of correlation between adhesion and J_0. The material then flows in response to the stress (creep portion, holding portion of adhesion test). After 10 s of holding during the adhesion test, the samples would not reach maximum compliance observed during creep, but the relative magnitudes of sample compliance based on PL would be the same. This region would correspond to the Kelvin-Voigt element within Burgers model, specifically λ_{ret} and J_1. A higher λ_{ret} at 10% PL implies more viscous behavior in the middle regime as retardation time is inversely related to elasticity (Ojijo et al., 2004). Additionally, λ_{ret} and J_1 both correlate strongly with adhesion, unlike the instantaneous elastic response J_0. This would also imply that long term viscous flow would not be happening yet, matching the lack of correlation between adhesion and η_N.

Keeping with this comparison, the recovery portion of the creep test would correspond to decompression during tack testing—so 10% PL may display more viscous behavior based on crp. The 10% PL sample contains more sucrose and corn syrup solids (i.e., mono- and disaccharides) that act as plasticizers and contribute to the viscous component (Saunders et al., 1992). This corresponded to greater adhesiveness than at higher PL, most notably higher adhesion to the high surface energy acrylic surfaces.

Lastly, to place these results in the context of oral processing, the omission of saliva and control of temperature in this study should be noted. It has been reported that the addition of saliva during texture profile analysis of fluids and semisolids only changed absolute values of adhesion and not trends among samples (Dunnewind et al., 2004). Moreover, strong correlations between pressure sensitive adhesion performed in the absence of saliva and sensory adhesiveness were reported for similar caramel samples (Wagoner et al., 2016). For simplicity in this study, all tests were performed at room temperature (22 ± 1C). Dynamic temperature changes occurring during oral processing would be expected to have a profound effect on rheological behavior, surface energy and, consequently, adhesion. Additionally, the formulations in this study had differing amounts of fat, so melting would also play a role in altering adhesion. As a result, future studies should consider how saliva may alter the surface energy relationship between food and oral surface, not to mention the material transformations that occur during oral processing that influence the temporal path of adhesion.

In conclusion, this study indicates that altering the proportion of fat and protein in a caramel formulation and the type of probe used for testing both influence pressure sensitive adhesion. Breaking down viscoelastic behavior into the components of a four-element Burger model was useful for predicting adhesion. Specifically, the components associated with retarded elasticity correlated strongly with adhesion. The results suggest two zones of adhesion based on formulation, one driven by surface energy relationships (most notably dispersive and total surface energy) and viscoelasticity, and the other driven solely by viscoelasticity.

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ETHICAL STATEMENTS

Conflict of Interest: The authors declare that they do not have any conflicts of interest.

Ethical Review: This study does not involve any human or animal testing.

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APPENDICES

APPENDIX A
DEVELOPMENT AND CHARACTERIZATION OF MILK PROTEIN CONCENTRATE MODEL SYSTEM

A1. Development of MPC model system

Stock MPC solutions were prepared by dispersing powder into deionized water at 600 rpm for 60 s, then in a 55 ˚C water bath for 120 min. Solutions were stirred using an overhead mixer (Heidolph RZR, Schwabach, Germany) with attached blade impeller (25 mm diam.) at 400 rpm to ensure complete hydration. pH 6.9 ± 0.1 adjusted with 1 N HCl or NaOH as required. The final composition of the model system containing MPC was 3.0% sucrose, 3.0% protein, 1.8% fat, and 1.4% lactose on a weight basis.

Food grade polysaccharides used in the study included: medium viscosity carboxymethyl cellulose (CMC) with a reported 250,000 g/mol molar mass and 0.7 degree of substitution (Sigma-Aldrich; St. Louis, MO, USA); λ–carrageenan reported as medium viscosity by the manufacturer (Danisco, Copenhagen, Denmark); agglomerated xanthan gum (TIC Gums, White Marsh, MD); enzymatically modified tapioca starch (Ingredion, Westchester, IL); high acyl gellan gum, kappa–carrageenan, and CT Blend® a proprietary blend of CMC, microcrystalline cellulose, and kappa–carrageenan (CP Kelco; San Diego, CA, USA). Food grade NaOH and HCl were purchased from Sigma-Aldrich and sucrose from a local grocery. Usage levels were formulated to match viscosity at 50 1/s (Fig. X).

The formation of a fluid gel directly with MPC required a separate hydration method for gellan gum. Gellan gum was first dispersed into water and mixed at 600 rpm with an overhead mixer in 85 ˚C water bath for 30 min. Solution was then moved to a 55 ˚C water bath and MPC and sucrose were added and hydrated for 120 min at 400 rpm. All other polysaccharides were prepared by dispersing powder into deionized water at 600 rpm in an 85 ˚C water bath for 30 min using the aforementioned overhead mixer with attached blade impeller. The CT blend sample was also mixed with Ultra Turrax (IKA Works, Inc., Wilmington, NC) at 12,000 rpm for 3 min to fully disperse the microcrystalline cellulose. Samples were then mixed with the necessary amount of MPC stock solution to reach desired final concentrations. All samples were cooled in an ice bath and stored overnight at 4 ˚C

before use. Samples for sensory analysis were used within 24 hours, all other samples included 200 ppm sodium azide as a microbial inhibitor and were used within 4 days.

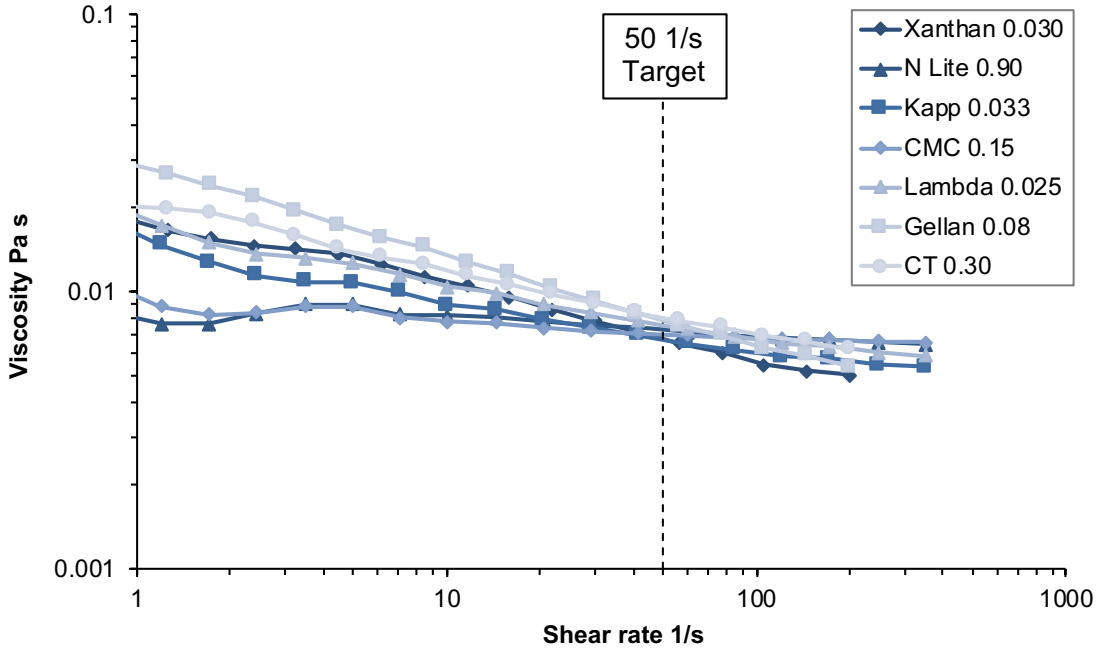

Figure A1. Flow profiles of MPC with various polysaccharides showing matched viscosity at 50 1/s.

A2. Confocal Laser Scanning Microscopy

Microstructural features of fluids were investigated using a Zeiss LSM 800 confocal laser scanning microscope (Carl Zeiss AG; Oberkochen, Germany). Samples were stained with a mixture of Nile Red (0.5% w/w in polyethylene glycol), Fast Green FCF (0.2% w/w in polyethylene glycol), and Fluorescent Brightener 28 to visualize dispersed fat, protein, and cellulose respectively. Samples were combined with stains at a 1:50 volume ratio of stain to sample and mixed in Eppendorf tubes for 10 min. The objective lens used were 40X and 63X oil immersion lens and a 1.8X digital zoom was applied in the software (Zeiss Zen Version 2.3). Excitation wavelengths were 488 nm and 640 nm. Digital image files were obtained at a resolution of 1024 by 1024 pixels. Images in the *xy* plane were obtained at a *z*-depth of at least 5 μm to avoid artifacts from the cover glass slip.

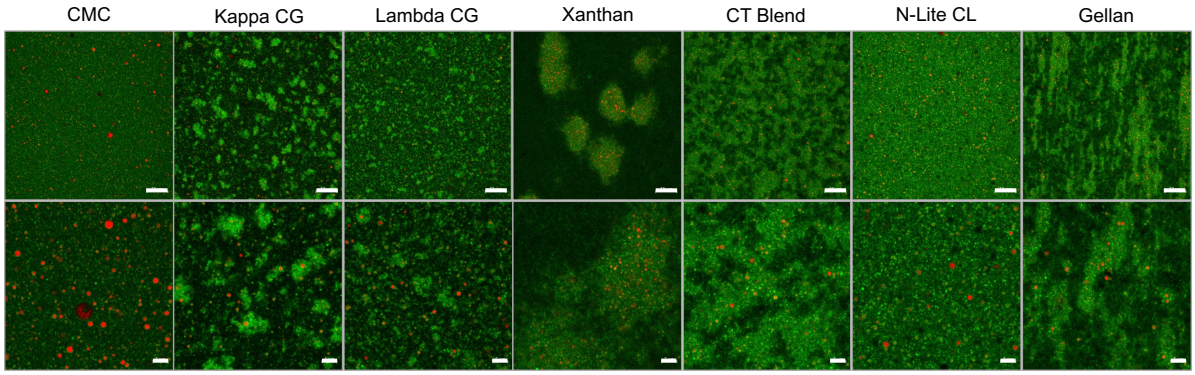

Figure A2. Confocal micrographs of MPC model system with different polysaccharides. Scale bar is 20 µm and 5 µm in the top and bottom rows, respectively.

Table A1. Correlations among sensory and rheological attributes of hydrocolloid solutions dispersed in reconstituted whole milk powder.

	Paste	Consistency	Creamy/Oily	Astringency	Mouthcoating	Sweet	Salt	Creamy	Cooked	Residual mouthcoating	Residual astringency	Visc 0.5	Visc 50	Visc 30000
Paste	1.00	0.98*	1.00*		0.94*			0.88*		0.93*	0.77		0.94*	
Consistency		1.00	0.97*	0.78	0.97*			0.84*		0.95*	0.77	0.76	0.98*	
Creamy/Oily			1.00		0.94*			0.89*		0.93*	0.81		0.92*	
Astringency				1.00	0.81					0.85	0.72		0.80	
Mouthcoating					1.00			0.75		0.96*	0.85*	0.84	0.99*	
Sweet						1.00								
Salt							1.00							
Creamy								1.00		0.85	0.72		0.77	
Cooked									1.00					
Residual mouthcoating										1.00	0.90*	0.75	0.95*	
Residual astringency											1.00	0.76	0.79	
Visc 0.5												1.00	0.84*	
Visc 50													1.00	
Visc 30,000														1.00

Only significant values shown ($p < 0.05$), asterisk indicates $p < 0.001$.

Table A2. Correlations among sensory and rheological attributes of hydrocolloid solutions dispersed in reconstituted skim milk powder.

	Paste	Consistency	Creamy oily	Astringency	Mouthcoating	Sweet	Salt	Creamy	Cooked	Cardboard	Residual mouthcoating	Residual astringency	Visc 0.5	Visc 50	Visc 30,000
Paste	1.00	0.99*	0.99*		0.98*						0.96*			0.97*	
Consistency		1.00	0.99*		0.98*			0.82			0.95*			0.97*	
Creamy oily			1.00		0.98*			0.82			0.95*			0.96*	
Astringency				1.00				0.88						0.99*	
Mouthcoating					1.00						0.95*			0.96*	
Sweet						1.00									
Salt							1.00								
Creamy								1.00						0.95*	
Cooked									1.00						
Cardboard										1.00					
Residual mouthcoating											1.00			0.92*	
Residual astringency												1.00		0.83	
Visc 0.5													1.00		
Visc 50														1.00	
Visc 30,000															1.00

Only significant values shown ($p < 0.05$), asterisk indicates $p < 0.001$.