Rheological Properties of Yogurt: Effects of Ingredients, Processing and Handling

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1 Introduction

Yogurt is one of the oldest and most popular fermented milk products consumed throughout the world. By definition, yogurt is a dairy product prepared by fermenting milk with the starter culture combination of *Lactobacillus delbrueckii* ssp. *bulgaricus* (*L. bulgaricus*) and *Streptococcus salivarius* ssp *thermophilus* (*S. thermophilus*). In the U.S., standards of identity specified in the U.S. Food and Drug Administration Code of Federal Regulations (CFR Title 21 part 131.200; USFDA 2018) dictate the optional base dairy ingredients to which the cultures are added: cream, milk, partially skimmed milk, or skim milk, used alone or in combination. This standard of identity also specifies other optional ingredients, nutritive carbohydrate sweeteners, and the minimum milk solids nonfat content (8.25%), minimum fat content (3.25%) and minimum titratable acidity (0.90%, expressed as lactic acid) required for the finished yogurt. The CFR provides flexibility in the use of flavoring ingredients, color additives, and stabilizers in yogurt. While yogurt by definition is made from whole milk, in today's U.S. market, "whole milk yogurt" is sold at a premium price. For decades, the predominant yogurt products available in the U.S. market were low-fat yogurt (required to contain not less 0.5% and not more than 2% fat) or nonfat yogurt (required to contain <0.5% fat) because of the perception that fat of all forms should be minimized in the human diet. Lite or light yogurts contain 1/3 fewer calories or 50% reduc-

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tion of fat compared to the regular yogurt. Additionally, they are typically sugar-free, and contain non-nutritive, high-intensity sweeteners.

The popularity of yogurt has increased in recent decades predominantly because of the health benefits associated with yogurt consumption. In the U.S., per capita consumption of yogurt increased from 6.5 lbs in 2000 to 13.7 lbs in 2017 (USDA-ERS 2018). The health benefits of yogurt are mainly attributed to the active starter and probiotic cultures used in the fermentation, along with the bioavailability of nine essential nutrients naturally present in yogurt (Chandan et al. 2017; Hill et al. 2017). The popularity of yogurt is maintained by the diversity of the yogurt products available in the market, ranging from plain to fruit-flavored and from drinkable to thick, spoonable Greek styles (Hill et al. 2017).

Along with health benefits, good sensory attributes are vital for the popularity of any yogurt product among consumers. Choices of culture type, ingredients in the formulation (such as addition of hydrocolloids, prebiotics, or high-intensity sweeteners), heat treatment of the yogurt mix, and fermentation temperatures used in manufacturing can significantly affect yogurt sensory attributes. Therefore, yogurt manufacturers and food scientists have conducted numerous scientific studies to understand the effects of the aforementioned parameters on the textural, rheological and other sensory attributes of yogurt. However, many publications in the food science literature highlight ingredients that “improve” yogurt texture, but do not set the context for what the “improvement” is. There is no single ideal yogurt body or texture: preferences for yogurt body and texture vary throughout the world. For instance, in China, yogurt is commonly consumed with a straw, but in the Middle East, a spoon is essential for yogurt consumption. Yogurt products from countries outside of the U.S. vary considerably in name, formulation, processing, and body and texture. For this reason, it is misleading when manuscripts include the word “better” or “improved” when describing the impact of particular ingredients or processing conditions on yogurt body or texture. It is essential, for this reason, for authors to describe changes in texture more explicitly and define “improvement” within the specific context of the expectations of the intended audience.

Although the market for non- and low-fat yogurt has been strong for many years, reducing or eliminating milkfat from the yogurt mix, which changes the rheological and textural properties of yogurt, continues to be of interest to consumers and processors. One negative outcome of removing milkfat from yogurt is an increase in syneresis, or free whey on the surface of yogurt. Research-based recommendations are available to counteract negative impacts of reduced milk fat in yogurt, including increased total solids in the yogurt mix by adding nonfat dry milk (NDM), skim milk powder (SMP), whey protein concentrates (WPCs) and isolates (WPIs), and caseinates; incorporation of stabilizers (such as gelatin, xanthan gum, guar gum, modified starch, etc.) into the formulation; and alterations of fermentation temperature and/or time (Nguyen et al. 2017; Teles and Flores 2007; Damin et al. 2009). Nevertheless, many challenges remain in developing stable lower-fat yogurt formulations with palatability similar to that of full-fat yogurts.

This chapter will begin with terminology related to rheological properties of yogurt, and review the effect of ingredients, processing, storage, and handling on yogurt rheological properties.

2 Terminology Related to Yogurt Rheological Properties

Broadly speaking, the term “rheological properties” encompasses concepts such as body, texture, mouthfeel, and microstructure. Although commonly used interchangeably, the terms body and texture are actually different. The term *body* generally refers to the overall physical structure of the bulk or majority of the yogurt. Some sub-terms that relate to yogurt body include, but are not limited to, firm, gel-like, lumpy, and weak (Tribby 2008). It is worth noting that the attribute “hardness”, though used commonly in literature regarding texture analysis of yogurt, is not entirely appropriate, since hardness is a measure of solid foods. Since yogurt is a semisolid food, the attribute “firmness” should be used.

The term *texture* should be used to more specifically refer to microstructure or arrangement of small constituent parts of yogurt. Szczesniak (2002) defined texture as the sensory and functional manifestation of the structural, mechanical and surface properties of foods detected through vision, hearing, touch and kinesthetics. Some sub-terms that relate to yogurt texture include, but are not limited to, chalky, grainy, mealy, grainy, and sandy.

Rheology is the study of flow and deformation of material. Hence, yogurt rheology is defined as how the yogurt gel flows and deforms under normal and/or tangential stresses. Key rheological concepts are outlined here, with more detail provided in Chapters “Introduction: Measuring Rheological Properties of Foods” and “LAOS (Large Amplitude Oscillatory Shear) Applications for Semisolid Foods”.

Shear stress is the force applied parallel or tangentially to the product cross-section. *Shear rate* is the rate at which shear is applied. *Shear strain* is the resultant deformation, or change in shape, size or volume of the material. *Yield stress* is the stress at which a material begins to deform. Rheological parameters used to describe yogurt consist of *(apparent) viscosity* (η), consistency index, *storage modulus* (G'), *loss modulus* (G'') and loss tangent (δ or G''/G') (Lubbers et al. 2004; Nguyen et al. 2017). Viscosity is the resistance to flow due to the internal friction of moving particles of fluid under force. While water is Newtonian (sheer stress directly proportional to shear rate), most yogurt products are non-Newtonian. Yogurt gels typically exhibit weak viscoelastic and *shear-thinning*, or *pseudoplastic* behavior (Damin et al. 2009; Lubbers et al. 2004; Sah et al. 2016), wherein the viscosity decreases with increased shear rate or stress. The increased shear rate changes the particle orientation of shear-thinning materials, causing a decrease of internal friction of the particles in the material. Shear stress and shear rate data of non-Newtonian fluids including yogurt can be analyzed using mathematical models. The Herschel-Bulkley model is the most commonly used model, but Ostwald, Steiger-Ory, Bingham, Ellis and Eyring models have also been used successfully (Dönmez et al. 2017; Hassan et al. 2003; Mathias et al. 2011).

On application of stress, viscoelastic materials store some deformation energy in their structure and lose some energy in the flow. The amount of energy stored versus dissipated is useful for determining various yogurt behaviors. For example, gelation in yogurt has been defined as the strain at which $G' > 1$ Pa. Yield stress (σ_{yield}) can be characterized as the point when shear stress value begins to decrease, which is observed by a decrease in the moduli values. Similarly, yield strain (τ_{yield}) can be defined as the corresponding strain value at that point (Lee and Lucey 2006). Low δ values indicate more solid-like, rubbery gels, while high δ values indicate more fluid-like, weak gels.

Numerous instrumental techniques are used to evaluate the body and texture of semisolid foods like yogurt, including but not limited to scanning electron microscopy, rheology, dynamic mechanical analysis, and tribology. With rheometry, microstructural changes that occur during gel formation or in the gel state can be revealed (Boubellouta et al. 2011). Oscillatory tests are particularly good at revealing mechanical spectra relating to G' and G'' (Chen and Stokes 2012; Stokes and Frith 2008; Conti-Silva et al. 2018). However, to ensure valid data that are comparable across multiple yogurt types and datasets, testing must be carried out under well-defined conditions and geometries. With spectroscopic methods, molecular structure changes in micelles throughout the acidification processes can be observed (Boubellouta et al. 2011). Synchronous fluorescence and infrared spectra are sensitive to changes in the micelle structure (phosphate dissolution, swelling of caseins) and interactions (casein with water, aggregation of particles), to help reveal interaction mechanisms (Boubellouta et al. 2011). Tribology, a newer technique, encompasses both rheological properties of the food and the surface properties of tongue and palate in relative motion (Nguyen et al. 2017; Conti-Silva et al. 2018). A fuller discussion of tribological testing of solid foods is presented in Chapter "Semisolid Food Tribology". Applications of techniques commonly used in the industry are discussed in Chapter "Rheological Testing for Semisolid Foods: Traditional Rheometry".

3 Styles of Yogurt

In the U.S. alone, there are over a dozen styles of yogurt commonly sold in the market, multiplied by the additional variations of each, resulting from different fat content (whole, low-fat, nonfat), sugar-free (light, lite), and flavor options. Brief explanations of several available yogurt products in the market are included in this section.

Cup-set yogurt is yogurt that is produced by blending ingredients, pasteurizing (with or without homogenizing), culturing, and filling of cups (or glass) with yogurt mix prior to incubation. When adequate fermentation has been completed (titratable acidity above 0.90%), cups are cooled. French yogurt is essentially cup-set yogurt. It may be flavored but does not contain fruit pieces.

Sundae-style yogurt is a cup-set yogurt typically called “fruit on the bottom” in supermarkets. After culturing, the yogurt mix is delivered on top of a fruit preparation that has been dropped into the bottom of cups. The sundae cups are subsequently incubated, then cooled after fermentation.

Swiss-style yogurt is also known as blended or stirred yogurt. After culturing, the yogurt mix is incubated in a large tank to the desired completion of fermentation. During cooling, flavoring, coloring, and fruits are gently blended into the curd. After blending, the yogurt cups are filled, sealed, and refrigerated. A newer option in the market, Australian yoghurt, is similar to Swiss-style yogurt, but is made with whole milk.

Custard-style yogurt is similar to Swiss-style except that it contains enough hydrocolloids to increase the firmness to create a custard-like consistency.

Cream-top yogurt is any style of yogurt that made from yogurt mix that has not been homogenized. During storage, the cream layer will rise to the top of the yogurt body, providing the cream top in the name.

In the case of drinkable yogurt, homogenization of the set curd occurs once the desired fermentation has taken place. Because the homogenization is performed at a higher shear rate, the final product has a lower viscosity than Swiss-style yogurt. Addition of flavoring and coloring may take place before or after the homogenization step. For a shelf-stable product, ultra-pasteurization and aseptic packaging are required steps. Kefir, a cousin of drinkable yogurt, is fermented by a diverse family of microorganisms, which include yeasts and lactic acid bacteria, contained in kefir grains.

Greek yogurt, also known as “strained” or Greek-style yogurt, is made by straining or centrifuging plain yogurt curd. It takes approximately 3 kg of regular yogurt to make 1 kg of Greek yogurt. Alternatively, dairy solids can be added to increase the protein content and thicken the yogurt. Greek-style yogurts contain approximately twice as much protein as conventional yogurt due to the removal of whey.

Icelandic yogurt is similar to Greek yogurt in that it is strained. However, it is typically made with nonfat yogurt mix and contains little to no sugar, making it quite sour to the American palate. Labneh, particularly popular in the Middle East, is similar to Greek yogurt in body, with a total solids content typically ranging from 23–25%, but the fat content is typically over 9% (Saleh et al. 2018). Dahi is very popular in India is similar to stirred yogurt. Varying from nonfat to full fat, dahi is prepared with multiple starter cultures, and the final product has higher acidity than stirred yogurts.

Whipped yogurt is made in a similar fashion as ice cream, wherein air is injected and entrapped in the structure. However, heat is not removed from the system so no freezing occurs. Compared to more traditional yogurts, whipped yogurts typically contain more sweeteners and hydrocolloids (typically gelatin) to maintain the foam structure.

In the U.S., frozen yogurt is a misnomer in that it is not pure yogurt that is frozen. Frozen yogurt is essentially low-fat ice cream mix that contains some (typically less than 15%) yogurt.

Incorporation of potentially probiotic bacteria during or after yogurt fermentation is common. Potentially probiotic microorganisms, sometimes listed on yogurt labels, include *Bifidobacterium bifidum*, *B. animalis*, *B. longum*, *Lactobacillus acidophilus*, *L. casei*, *L. plantarum*, *L. reuteri*, and *L. rhamnosus* (Chandan 2006; Hill et al. 2017). For a yogurt to be truly probiotic, however, rigorous testing must be conducted to determine the live cell count over the shelf life of the product and demonstrate the beneficial effects on the human gastrointestinal tract

4 Rheological Changes in Yogurt Resulting from Ingredients

In its most basic form, yogurt is composed of pasteurized milk and cultures, yet most yogurt in the U.S. contains more than those two ingredients. Each ingredient plays a critical role in the rheological properties and consumer experience with yogurt. The roles of these ingredients, in six broad categories, are summarized in the following subsections. Overall, the information in this section emphasizes the need for careful consideration of all ingredients in a yogurt formulation, including milk source, byproducts of microbial cultures, functional and flavoring ingredients, and added micronutrients.

4.1 Use of Milk from Different Species

Considering the importance of mammals other than cows around the world, yogurt made from the milk of buffalos, camels and small ruminants should not be overlooked. For example, yogurt made with goat milk forms softer curds than yogurt made from bovine milk because of the naturally lower amount of alpha$_{s1}$-casein in goat milk. Similarly, camel milk produces a weak yogurt body (Abou-Soliman et al. 2017). Sheep milk, on the other hand, has a higher solids content and produces yogurt with a more firm and resilient curd structure, especially compared to yogurt made from goat milk (Gursel et al. 2016).

Gursel et al. (2016) manufactured goat milk yogurts with fortification of 2% (w/v) skim goat milk powder (SGMP), sodium caseinate (NaCN), WPC, WPI, or yogurt texture improver and stored it 21 d at 5 °C. Compared with goat milk yogurt made by using SGMP, the other yogurts had higher protein content and lower acidity values. Yogurts fortified with either NaCN or yogurt texture improver had more compact structure and lower syneresis than yogurt fortified with WPC. Using WPI caused the firmest yogurt body and higher syneresis. Acetaldehyde and ethanol formation increased with the incorporation of WPI, WPC, or yogurt texture improver into the yogurt base. Counts of *S. thermophilus* were higher than counts of *L. bul-*

garicus, possibly due to a stimulatory effect of milk protein-based ingredients other than SGMP on the growth of *S. thermophilus*. Yogurt with NaCN received the highest body and texture scores from trained Turkish panelists.

Costa et al. (2015) investigated the impact of cupuassu (*Theobroma grandiflorum*); a fiber-rich acidic fruit similar to cacao that is cultivated in Brazil, Colombia, Bolivia, and Peru; to modify goat milk yogurt body. Cupuassu pulp, probiotic (*Lactobacillus acidophilus* LA-5), and prebiotic (inulin) ingredients were used to make yogurts called "natural", "probiotic", "prebiotic", "symbiotic" (probiotics plus inulin), and "probiotic with cupuassu". All yogurt samples underwent gradual decreases in pH until 7–14 d of refrigerated storage, but the probiotic bacteria remained viable (≥7 log cfu/mL) throughout 28 d of refrigerated storage. When used alone, addition of inulin and cupuassu increased the apparent viscosity of goat milk yogurts for up to 21 d storage compared to the viscosity of other yogurts. However, by day 28, no differences in yogurt viscosity were apparent. Additionally, at the end of storage, the consistency was higher in the yogurts with inulin (prebiotic and symbiotic), but no other meaningful differences were seen throughout storage, including no differences in firmness at any time. While the authors concluded that "cupuassu is an important technological strategy for the dairy goat industry", that appears to be an overstatement based upon closer inspection of the results.

Although camel milk is an important food source worldwide, it is not successful in yogurt production, in part explained by the large casein micelles, little to no β-lactoglobulin (β-lg), and small milk fat globules (Abou-Soliman et al. 2017). There has been some effort to improve camel milk yogurt with SMP (Salih and Hamid 2013) or the addition of hydrocolloids and stabilizers (Al-Zoreky and Al-Otaibi 2015) with little success. Abou-Soliman et al. (2017) investigated the impact of microbial transglutaminase (0.4% concentration) with and without bovine SMP, WPC, or β-lg on physicochemical, rheological, microstructural, and sensory properties of camel milk yogurt during 15 d storage. Fortification of camel milk with dairy ingredients alone, without microbial transglutaminase, did not set. Microbial transglutaminase treatment yielded yogurt, and the addition of bovine powders increased the protein matrix in the gel microstructure. The highest firmness values were obtained for samples made with SMP-fortified milk, and the lowest was for WPC-fortified milk. Microbial transglutaminase not only improved camel yogurt firmness, but increased the viscosity and water-holding capacity of the yogurts, showing promise for the ingredient for camel milk yogurt applications.

4.2 Optional Additional Dairy Ingredients

Common optional additional dairy ingredients used in yogurt base formulations include NFDM or SMP, caseinates and various WPCs. It is logical to think that increasing solids in yogurt yields an increase in yogurt firmness. However, that is not always the case: both the type of solids added and processing conditions impact yogurt body and texture. Additionally, increased solids may induce other changes to yogurt that may not be acceptable to the consumer (e.g., chalky or grainy). Careful selection of all ingredients used in yogurt, as well as processing and fermentation conditions, is essential.

It has long been known that the changes in chemical composition of a yogurt base due to protein fortification influence the rheological and physical properties of yogurt (Peng et al. 2009; Lee and Lucey 2010). Several studies have investigated the effects of fortification of milk with WPCs and caseinates on the physical properties of yogurt (Remeuf et al. 2003; Isleten and Karagul-Yuceer 2006; Peng et al. 2009; Marafon et al. 2011). Heat treatment of milk fortified with WPC induces crosslinking within the gel network, which results in a dense yogurt structure and increased yogurt viscosity and water holding capacity (Remeuf et al. 2003). Fortification with WPC has been shown to decrease syneresis and increase viscosity and firmness compared to fortification with SMP or whey powder. This was attributed to the disulfide bridges formed between denatured whey proteins in WPC with the casein micelles in milk (Bhullar et al. 2002; Marafon et al. 2011). Casein interactions play an important role in the textural properties of yogurt (Peng et al. 2009). For instance, a low casein content is believed to yield a more open gel structure, making the coagulum network more sensitive to syneresis (González-Martınez et al. 2002; Zhao et al. 2016). The non-protein nitrogen content of milk powders was shown to negatively influence the viscosity and thickness of fortified yogurt, which was highest for yogurt fortified with milk protein concentrate (MPC), followed by SMP, and lowest for casein hydrolysate (Sodini et al. 2002). The addition of increasing levels of β-lg has been reported to cause marked increases in storage modulus compared with α-lactalbumin (α-la), with some differences in behavior among different β-lg variants (Graveland-Bikker and Anema 2003). The amount of fat and distribution of fat was shown to improve the firmness and texture of yogurt fortified with either WPC, whey powder, or SMP (Bhullar et al. 2002).

Yu et al. (2016) studied the effect of adding instant NFDM on the physical properties and microstructure of yogurt. The physical properties of fat-free yogurt, fat-free yogurt with NFDM, whole-fat yogurt, and whole-fat yogurt with NFDM were analyzed using rheometry and imaging techniques. Not surprisingly, the two yogurts that incorporated NFDM had higher consistency coefficient, storage modulus, yield stress, and firmness. Nuclear magnetic resonance (NMR) and brightfield microscope images showed that NFDM contributed positively to strengthening the physical structure, thus altering the mechanical properties of the yogurts.

Marafon et al. (2011) evaluated the quality of Swiss-style probiotic nonfat yogurt fortified by partially replacing SMP with WPC and sodium caseinate (NaCN) compared with non-fortified yogurt. Yogurt rheological properties were measured using dynamic oscillation over a 28 d storage period. Higher G' and G'' values and more homogeneous microstructures were found in the fortified yogurts, and higher gel strength was maintained in these yogurts during storage. Neither the acidification profile nor viable counts of probiotic bacteria were affected by supplementation of the solids in the yogurt base. A sensory study with 120 untrained participants was conducted on the appearance, flavor, and texture of the yogurts using a 9-point hedonic scale. The results revealed that yogurt made with fortification of milk with WPC and NaCN had acceptable appearance, acidic taste and firmer consistency throughout their shelf life compared to yogurt made with no fortification (Marafon et al. 2011).

In an effort to increase the protein content and consistency, while imparting creaminess to yogurt, Morell et al. (2015) prepared yogurts with SMP and WPI, as well as a control without extra protein. Three additional samples were prepared by adding 2% of a physically modified starch to each. A controlled-stress rheometer was used to characterize the flow and viscoelastic properties of the samples before and after *in vitro* oral digestion, and their microstructure was observed with light microscopy and low-temperature scanning electron microscopy. Before *in vitro* oral digestion, samples with SMP showed denser areas than the control yogurt; in samples with WPI, two protein networks could be distinguished. In the samples with added starch, starch granules were embedded in the protein networks. After *in vitro* oral digestion, the protein tended to aggregate; the starch granules maintained their structure, indicating that they were not broken down by the saliva. All samples showed pseudoplastic behavior, as well as $G' > G''$, describing a weak gel structure with elastic characteristics. While samples with WPI exhibited the highest consistency index, yogurts made with starch showed higher viscosity than those without starch because the starch acted as fillers, strengthening the protein network (Morell et al. 2015).

Zhao et al. (2016) delved more deeply into the effect of casein to whey protein ratios (4:1, 3:1, 2:1, and 1:1) on gelation properties and microstructure of low-fat yogurt made with reconstituted skim milk with or without addition of WPC. The rheological properties of the low-fat yogurts were evaluated using a Universal Dynamic Spectrometer. The microstructure (measured by confocal scanning laser microscopy) became more compact with smaller pores as the ratio of casein to whey proteins decreased. When the ratio of casein to whey proteins was 2:1 or 1:1, the yogurt coagulum showed higher G' and greater yield stress, with more compact cross-linking and smaller pores than when casein levels were higher. In addition, when more SMP was replaced by WPC, a greater number of disulfide bonds were formed and hydrophobic interactions increased during heat treatment, tightening the microstructure of the final yogurt and increasing its firmness (Zhao et al. 2016).

In an effort to develop and optimize an alternative make procedure for Greek-style yogurt to reduce the amount of acid whey produced, Bong and Moraru (2014) incorporated micellar casein concentrate (MCC) into the base instead of using a straining step to increase yogurt solids. Two MCC preparations with 58 and 88% total protein were used to fortify yogurt base to 9.80% (w/w) protein, with strained Greek-style yogurt of similar protein content as the control. Regardless of inoculation level, the acidification rate was faster for the MCC-fortified Greek-style yogurt than for the control, which was attributed to the higher non-protein nitrogen content in the MCC-fortified milk. Steady shear rate rheological analysis indicated shear-thinning behavior for all Greek-style yogurt samples. Dynamic rheological analysis at 5 °C showed a weak frequency dependency of G' and G'' for all Greek-style yogurt samples, with $G' > G''$, indicating a weak gel structure. The lower water-holding capacity for the MCC-fortified Greek-style yogurt compared with the control was attributed to lower serum protein content in the MCC-fortified Greek-style yogurt. Despite some differences in the physicochemical characteristics compared to Greek-style yogurt manufactured by straining, the alternative process was considered a feasible alternative to the traditional Greek-style yogurt straining, with environmental and possibly financial benefits to the dairy industry (Bong and Moraru 2014).

The form of whey protein used as an ingredient also has an impact on rheological properties of fermented beverages. Dimitreli et al. (2013) studied the impact of heat treatment and whey protein addition on the fermentation time and the rheological properties of kefir using a pneumatic tube viscometer of novel design. Heat treatment of the milk was made prior to or after addition of various levels of WPC (to yield native or denatured whey protein, respectively). Increasing WPC concentration increased lactic acid concentration and reduced fermentation time. The flow curves of the samples demonstrated kefir's pseudoplastic fluid behavior. The apparent viscosity of kefir samples increased with increasing WPC concentration, but denatured whey proteins yielded higher consistency index values (higher apparent viscosity and lower flow behavior index values) compared with native whey proteins.

A novel ingredient that may be considered for yogurt applications in the future to help in the clean label movement may be CO_2-treated milk protein concentrate powder (TMPC). Meletharayil et al. (2018) mixed TMPC80 with NFDM in different ratios in the manufacture of acid gels with 4% (w/w) protein and 12% (w/w) total solids. Dispersions were adjusted to pH 6.5, followed by heat treatment at 90 °C for 10 min, then glucono-δ-lactone (GDL) was added and samples were incubated at 30 °C until pH 4.5 was reached (about 4 h). GDL levels were adjusted to compensate for the lower buffering capacity of samples with higher proportions of TMPC80 (attributable to the depletion of buffering minerals from the serum and micellar phase during preparation of TMPC80). When the proportion of protein contributed by TMPC80 was increased from 0% to 60%, gelation pH, gel porosity decreased and water-holding capacity and the G' of the gels at pH 4.5 increased. The authors concluded that because of decreased buffering and reduced need for hydrocolloids, the productive capacity of yogurt manufacturing plants may be improved by partial substitution of NFDM with TMPC80.

4.3 Addition of Hydrocolloids

It is common practice to add hydrocolloids of various forms to the yogurt base to bind water, thicken the final product, and reduce syneresis, which some consumers consider objectionable, during storage and shipping. It is impractical to provide a complete list of hydrocolloids that have been tested in yogurt applications, as dozens of hydrocolloids and hydrocolloid blends have been used, and more are being developed each day. Instead, an overview of some of the recent research with hydrocolloids in yogurt is summarized in this section. All of these studies highlight the impact of different hydrocolloids on yogurt rheology and texture, emphasizing the need for careful selection of hydrocolloids in the yogurt system.

In the U.S., one of the most traditional choices of hydrocolloids for yogurt applications is gelatin. Being a standard, and because it is an animal-based ingredient that is not desirable to some consumers, other hydrocolloids are often compared to gelatin. Pang et al. (2015) studied the effects of polysaccharides with different ionic charge on rheology, microstructure, texture and water-holding capacity of acid milk gels compared to gelatin. Similar to gelatin, starch (neutral) and xanthan gum (anionic) did not prevent milk gelation in the first 30 min of acidification, even at high concentrations. In contrast, two neutral polysaccharides, guar gum ($\geq$0.05%) and locust bean gum ($\geq$0.1%), inhibited milk gelation from the beginning of acidification. Carrageenan, another anionic polysaccharide, induced earlier milk gelation at low concentration ($\leq$0.05%), but inhibited gelation entirely at high concentration (0.2%). The highest water-holding capacity was seen with gelatin inclusion. Xanthan gum and starch were more similar to gelatin in their effect on acid milk gels compared to guar gum, locust bean gum and carrageenan.

Later, Pang et al. (2016) combined gelling polysaccharides (xanthan/locust bean gum, carrageenan, and starch) and milk proteins (WPI, NaCN, and SMP) in an effort to use them to replace gelatin in acid milk gels. Gels with added xanthan/locust bean gum alone showed rheological and microstructural properties similar to gels with gelatin. Similar to the effect of adding gelatin, milk protein fortification enhanced water-holding capacity of the gels, with WPI being the most effective. Gels with combinations of polysaccharides (except carrageenan) and WPI were stronger and had higher water-holding capacity than gels with no stabilizer. In yogurt, the combination of WPI and xanthan/locust bean gum produced similar effects on consistency, pseudoplasticity and apparent viscosity as with gelatin. In ranking tests with 38 untrained panelists, yogurt with WPI and xanthan/locust bean gum had higher thickness and stickiness than with gelatin, and lower smoothness than with gelatin.

Hematyar et al. (2012) prepared yogurt by supplementing yogurt mixes with 0.01 and 0.005% xanthan or carrageenan and evaluated yogurt characteristics during storage (4 °C at 10 d). Viscosity of supplemented yogurts was greater than non-supplemented yogurt, and viscosity increased during storage in all supplemented yogurts but decreased in the yogurt without supplements. The increased viscosity of the carrageenan-supplemented yogurts was explained by the presence of electrostatic interactions of anionic carrageenan and the net positively-charged casein micelles, whereas the viscosity increase in the xanthan-supplemented yogurt was explained by the increased viscosity of the continuous phase due to the addition of xanthan gum, which is an excellent thickening agent.

In an effort to evaluate resistant starch from maize in a dairy application, Lobato-Calleros et al. (2014) prepared three reduced-fat stirred yogurts from reconstituted milk (12.5 g/L of milk fat) with added native maize starch, and chemically modified maize or tapioca starches (10 g/L). The chemical composition, syneresis, rotational and oscillatory shear rheological properties and syneresis of the reduced-fat yogurts were evaluated and compared with those of a full-fat control yogurt (25 g/L of milk fat) without starch. The control yogurt exhibited lower apparent viscosity-shear rate profiles and dynamic viscoelastic moduli, but higher syneresis than the reduced-fat yogurts and all exhibited weak gel microstructural networks. The reduced-fat yogurts showed little variation in their flow and viscoelastic properties over 15 d of storage time. Overall, the addition of native or chemically modified starches contributed to the formation of stable reduced-fat yogurts.

Saleh et al. (2018) investigated the effects of hydrocolloids when used as fat replacers on the physicochemical properties of labneh compared to a 16% fat control. Four sets of combinations of carboxymethylcellulose (CMC), gum arabic, κ-carrageenan, and xanthan gum were used in a three-factor mixture response surface methodology. The maximum contribution of each individual hydrocolloid was set at 5% for xanthan gum, CMC, and gum arabic, and 1% for κ-carrageenan. Xanthan gum and κ-carrageenan increased labneh water-holding capacity, while CMC decreased water-holding capacity. Most samples showed shear-thinning behavior; penetration force increased when more than two hydrocolloid types were used, which was attributed to the formation of a 3-dimensional network that physically retained water. Viscosity and penetration results were hypothesized to be related to water-holding capacity. The water-holding capacity was highest in labneh with xanthan gum, followed by gum arabic, κ-carrageenan, then CMC. They found that xanthan gum exhibited an antagonistic effect with CMC that led to the decrease in water-holding capacity. Consumer testing (n = 40) revealed that an acceptable reduced-fat labneh could be produced by using hydrocolloids as fat replacers, which is remarkable since labneh typically contains more than 9% fat.

In recent years, some authors have used crosslinked acetylated starch to improve freeze–thaw stability of starches. Crosslinking reinforces hydrogen bonds in starch granules; acetylation induces structural reorganization and increases swelling power of granules (Tang et al. 2018). Cui et al. (2014) evaluated the effect of crosslinked acetylated cassava starch on set yogurt by investigating yogurt flowability, viscoelasticity, zeta potential, conductivity, and microstructure. The results indicated that the stability and viscoelastic moduli values of the set yogurt system increased with increased concentrations of crosslinked acetylated starch. Set yogurt systems with added crosslinked acetylated starch also exhibited shear-thinning behavior. SEM micrographs demonstrated that the microstructures were mainly composed of a casein network that was strengthened by adding crosslinked acetylated starch. The authors hypothesized that the starch adsorbed onto the surface of the casein micelles, preventing flocculation of the casein micelles by electrostatic adhesion, steric stabilization, and osmotic effects.

4.4 Use of Exopolysaccharide-Producing Cultures

Exopolysaccharides (EPS) are large polymeric carbohydrates. Yogurt that contains EPS may be described with a variety of terms, such as slimy, sticky, thick, lubricating, mouth-coating, ropy, or even "snotty", depending on the type of EPS produced. There are several types of EPS. Capsular EPS is synthesized inside the cell, and when released from the cell, remains attached to the exterior of the cell in the form of a capsule. In contrast, "ropy" EPS is excreted directly into the medium as a free-floating polysaccharide (Low et al. 1998; Cerning 1995; De Vuyst and Degeest

1999; Broadbent et al. 2003; Khanal and Lucey 2017). In some cases, bacteria can produce both forms of EPS.

Ropy describes how the yogurt strings up to follow a lifted spoon after the spoon is lifted 3–6 cm from the surface of stirred yogurt. EPS-producing or "ropy cultures" are often used, intentionally, to provide body to clean-label yogurt. Because capsular and ropy EPS possess high water-binding ability, the use of EPS-producing starter cultures helps decrease the level of whey separation in set yogurt (Wacher-Rodarte et al. 1993; Hassan et al. 1996; Jaros et al. 2002). Ropy cultures are influenced by pH, temperature, supplementation with WPC, and competition by other cultures in the yogurt base (Zisu and Shah 2003).

Ramchandran and Shah (2009) studied the effect of EPS- and non-EPS-producing *S. thermophilus* cultures on the rheological and textural properties of yogurt supplemented with inulin during 28 d storage at 4 °C. Results showed no significant changes in the yogurt firmness during storage. On the other hand, EPS-containing yogurts had lower firmness and G' and G'' values during storage compared to the non-EPS-containing yogurts. Ramchandran and Shah (2009) suggested that the interference of inulin between casein micelles could result in weaker yogurt gels. Moreover, the presence of void spaces around the EPS-producing bacteria could also contribute to the weaker yogurt structures. All yogurts showed $G' > G''$ throughout storage, confirming a weak gel microstructure. The lower G' values of EPS-containing yogurts indicated a less rigid gel microstructure compared to the non-EPS-containing yogurt. Overall, both EPS-containing and non-EPS-containing yogurts had similar loss tangent values during storage. However, the loss tangent did decrease in both types of yogurt during storage, indicating development of a more solid-like gel. All yogurts were showed pseudoplastic flow behavior at all timepoints and fit well to the Herschel-Bulkley model. Overall, yield stress increased significantly for both EPS-containing yogurts and non-EPS-containing yogurts, and yield stress was greater for non-EPS yogurts compared to EPS-containing yogurts. Ramchandran and Shah (2009) also reported a strong, significant correlation between firmness of EPS-containing yogurt ($r = 0.86$) and yield stress of non-EPS-containing yogurt ($r = 0.83$). The hysteresis loop area of EPS-containing yogurts and non-EPS-containing yogurts indicated thixotropic behavior of yogurt gels. The overall hysteresis loop area of both yogurts increased during storage and hysteresis area of EPS-containing yogurts was less than non-EPS-containing yogurt during storage. However, the thixotropic nature of yogurt gels was dependent on the type of EPS-producing strain (Ramchandran and Shah 2009; Amatayakul et al. 2006).

Later, Ramchandran and Shah (2010) studied the effect of addition of probiotics (*L. acidophilus*, *L. casei* and *B. longum*) with inulin on rheological properties of yogurt prepared with EPS- and non-EPS-producing *S. thermophilus*. The overall firmness of both yogurts increased over the 28 d storage time. Similar to their previous work, the firmness of non-EPS-containing yogurt was greater than EPS-containing yogurt at the end of storage. Similar trends were seen for yield stress and thixotropic behaviors.

Prasanna et al. (2013) prepared yogurts with starter cultures and EPS-producing *Bifidobacterium longum* ssp. *infantis* or *B. infantis* NCIMB 702205 and reported that G' values and firmness of yogurt increased during 28 d of storage at 4 °C. These increases over time were thought to be due to the rearrangement of proteins that formed the gel and increased interaction between the protein–protein and EPS–protein networks. On the other hand, Liu et al. (2017) prepared yogurts using EPS-producing *S. thermophilus* S3.3 with EPS-producing *L. bulgaricus* LTM or mutant EPS-producing *L. bulgaricus* LTM at three different fermentation temperatures (30, 37, and 42 °C), and evaluated these yogurts periodically throughout 21 d of storage at 4 °C. Liu et al. (2017) reported that yogurts prepared from the two EPS-producing *L. bulgaricus* strains differed in firmness, consistency, and cohesiveness. The differences in theses textural properties did not follow any specific trend. However, yogurts fermented at higher temperatures (37 and 42 °C) had greater firmness, consistency, and cohesiveness throughout storage compared with the yogurt fermented at 30 °C. In these systems, the milk proteins crosslinked with EPS, resulting in enhanced viscosity. Interestingly, Liu et al. (2017) reported that G' values increased during storage and hypothesized this result was related to the number of protein–EPS crosslinks. However, the specific effect of EPS on yogurt rheology varies with type of EPS-producing culture(s) and the type and quantity of EPS produced (Liu et al. 2017; Rawson and Marshall 1997).

Khanal and Lucey (2017) set out to understand if different strains of EPS-producing strains of *S. thermophilus* produce different yield and molar mass of EPS under the same conditions. Milk samples were analyzed for EPS concentration every 30 min during a fermentation period of 270 min (final pH 4.5) by using a modified quantification method. Both strains appeared to start producing significant amounts of EPS after ~150 min of fermentation, which corresponded to pH ~5.3, close to the gelation point. During the remainder of the fermentation process (150–270 min), the EPS concentration from the two strains significantly increased and was estimated to represent ~60% of the total EPS added to milk. In addition, distinct differences in rheological properties were seen between yogurts containing each of the two strains. At the end of fermentation, yogurts containing one strain produced weaker gels and higher maximum loss tangent values that occurred earlier during fermentation compared to yogurt made with the other strain. The differences were attributed to differences in chemical structures and molecular mass of the EPS produced by the two *S. thermophilus* strains since the fermentation conditions were identical (Khanal and Lucey 2017). These findings underline the importance of careful selection of culture strains to obtain desired results.

4.5 Addition of Fruit, Vegetables or Herbs

The addition of fruit to yogurt contributes to the abundance of options for consumers. Flavored and fruit yogurts are popular yogurts in the market. Strawberry, apple, mango, cherry, blueberry and banana are the most common fruits used in commerical yogurt in the U.S. Low-fat yogurt with fruits are particularly popular. Although addition of vegetables and herbs are being studied in yogurt preparation, these ingredients have not gained as much favor, at least in the U.S., and shelf life issues have not been fully addressed.

Lubbers et al. (2004) reported increased consistency index and apparent viscosity of stirred fruit yogurt (prepared from strawberry pulp) during 28 d of storage at 10 °C. Both consistency index and apparent viscosity increased during storage, although no significant change in flow behavior index was noticed due to fruit prep. Lubbers et al. (2004) attributed the changes in yogurt rheological behaviors during storage to the production of lactic acid and EPS from residual microbial activity, which would reinforce the protein network. On the other hand, Sengul et al. (2014) reported that yogurts supplemented with 8, 12, or 16% strawberry pulp did not exhibit an overall significant change in apparent viscosity. As viscosity depends on the yogurt acidity, the effects of fruit supplementation of yogurt depend on the preparation and physicochemical properties of the fruit supplement and its ultimate impact on yogurt acidity.

Dabija et al. (2018) studied the rheological properties of yogurt supplemented with herb extracts of thistle, hawthorn, sage, and marjoram at concentrations of 0.25–1.0% w/w. All yogurts showed thixotropic and weak gel characteristics. The overall viscosity of yogurt made without herb supplements was greater than that of the herb-supplemented yogurts on d 1; however, on d 28, the herb-supplemented yogurts had higher viscosity than the yogurt made without herb supplements. For all yogurts, $G' > G''$, and at day 28, the viscoelastic moduli values were greater in herb-supplemented yogurts compared with yogurts made without herb supplements, indicating a stronger gel network.

4.6 Yogurt with Other Functional Ingredients

A variety of functional ingredients have added to yogurt with efforts to improve nutritional properties for consumers and/or for select microorganisms. For instance, numerous dietary fibers are symbiotic because they are non-digestible and thus non-caloric to humans but promote the survival of probiotic microorganisms in the host gastrointestinal tract, and in turn benefit the host. Prebiotics are sometimes added to yogurt mix to support the growth and viability of starter and probiotic bacteria during fermentation and refrigerated storage. Functional ingredients, such as fibers, phytosterols and plant extracts, are becoming common yogurt components (Izadi et al. 2015; Sah et al. 2016). It is important that the type and quantity of additives used in the yogurt formulation not negatively impact the final sensory quality or safety attributes of the yogurt products. Yet inclusion of such ingredients can have direct impact on the body and texture of the yogurt. For instance, yogurt viscoelastic behaviors or apparent viscosity can be increased two- to three-fold by adding polysaccharides or enzymes (lactoperoxidase, protease, and transglutaminase) to the yogurt base, allowing new crosslinks to form in the gel network and enhancing gel rigidity and water-holding capacity (Zhao et al. 2016). This section elaborates on some of the recent research in the area.

Hoppert et al. (2013) analyzed the responses of a large number of young educated consumers (n = 704) on standard or 30% reduced-sugar vanilla yogurt enriched with inulin or with inulin combined with a grain mixture, a milled mixture of flakes, or a combination of grains and milled flakes (>1.5 g fiber per 100 kcal). Hedonic acceptability and Just About Right testing were conducted with 88 panelists judging each of six yogurts. Overall, sugar content was found to be the primary influence on yogurt acceptability, adapting the flavoring concentration might be an appropriate tool to mask sugar reduction, and the size of incorporated fiber should be considered in product optimization to minimize cereal flavor and appearance of fiber particles.

Sah et al. (2016) studied the effect of including 1% w/v inulin or pineapple peel powders in yogurt formulations on the physicochemical, textural, and rheological properties of set-type yogurt with or without probiotics (*L. acidophilus*, *L. casei* and *L. paracasei*) during 28 d of storage at 4 °C. All yogurts demonstrated weak gel and non-Newtonian behaviors. Sah et al. (2016) reported that the firmness of yogurts without added fiber increased during storage, which was attributed to the decrease in yogurt pH (from 4.49 on d 1 to 4.29 on d 28) during storage, resulting in the shrinkage of gel structure and elevation of gel strength. However, yogurts prepared with fiber had lower firmness throughout storage compared to yogurts without fiber. These results indicate poor compatibility between the milk proteins and inulin and pineapple peel fibers. Yogurt G' values increased during storage, which was attributed to gel shrinkage, increased gel strength, increased elastic-type behavior, or a combination of these factors. Although addition of probiotics and inulin did not affect the G' values during storage, yogurts containing pineapple peel powder did have lower G' values throughout storage, implying that pineapple peel powder fortification resulted in weaker gels with less elastic-type behavior. Further, Sah et al. (2016) reported that the loss tangent values decreased during storage, which was interpreted as a rearrangement of gel structure that improved gel elasticity. Although the apparent viscosity of the non-supplemented yogurts remained consistent during storage, the apparent viscosity of the supplemented yogurts increased. The overall yield stress, calculated using the Herschel-Bulkley model, increased during storage for the pineapple peel powder-supplemented yogurt, although this sample had the lowest yield stress throughout storage. Micrographs of yogurt structures generated by Sah et al. (2016) showed a more densely packed casein network after storage, which may explain the higher G' values at the end of the storage.

Bakirci et al. (2017) studied the effect of supplementing low-fat set-style yogurt with 0.5, 1.0, or 1.5% pumpkin fiber on the rheological properties during 14 d of storage at 4 °C. The apparent viscosity of yogurt increased with increasing pumpkin fiber concentration. Moreover, the viscosity of all pumpkin fiber-supplemented yogurts was greater than that of the non-supplemented yogurt throughout storage. As pumpkin is a good source of pectin, the increased viscosity was attributed to the pectin's contribution to the water-binding ability of the supplemented yogurts. Bakirci et al. (2017) reported that all yogurts exhibited a weak gel microstructure and elastic-dominant behavior ($G' > G''$) throughout storage. Both G' and G'' were greater in pumpkin fiber-supplemented yogurts compared to the non-supplemented yogurt. Bakirci et al. (2017) attributed the increased in viscoelastic moduli values to the increased interactions between the casein micelles and the pectin from pumpkin fibers. Scanning electron microscope images of yogurt gels showed that the pumpkin fibers filled the void spaces between the casein aggregates, creating a denser gel structure compared to non-supplemented yogurt, which showed larger voids. Bakirci et al. (2017) concluded that using pumpkin fibers as an ingredient could improve the quality and textural and nutritional properties of reduced-fat yogurts.

Knowing that interactions between polyphenols and proteins are based on weak hydrophobic, van der Waals, hydrogen bridge-binding, and ionic interactions formed between amino acid side chains and polyphenol aromatic rings, Dönmez et al. (2017) studied the effect of added green coffee powder and green tea powder on the syneresis and flow behaviors of set yogurts. Adding green coffee powder at 1 or 2% concentration decreased syneresis. However, in comparison to the control, green tea powder decreased syneresis when added at 0.02%, but increased syneresis when added at 2%. Herschel-Bulkley model parameters indicated that the consistency coefficient of the control yogurt was lower than that of green coffee powder-containing yogurts for up to 14 d, but was higher after 21 d of storage. Consistency coefficients of yogurts made with 0.01 or 0.02% green tea powder were higher than those of the controls, but consistency coefficients of yogurts made with 1 or 2% green tea powder (1 or 2%) were lower than those of the controls. Dönmez et al. (2017) hypothesized that the polyphenols in green coffee and green tea interacted with casein micelles, and differences in the polyphenol profiles of the powders played a role in the observed differences of the yogurts.

Mudgil et al. (2017) studied the effect of partially hydrolyzed guar gum (a dietary fiber) level (1–5%), culture level (1.5–3.5%), and incubation time (4–8 h) on yogurt texture profile analysis behaviors. Fortification of yogurt with partially hydrolyzed guar gum or culture beyond 2.5% decreased yogurt firmness. Partially hydrolyzed guar gum fortification also decreased gumminess and increased the adhesiveness, cohesiveness, and springiness of yogurt.

Santillan-Urquiza et al. (2017) studied the effect of fortification of set-type yogurt with iron oxide, zinc oxide, and calcium phosphate, added as inulin-coated nanoparticles or microparticles, on the physicochemical and rheological properties of yogurt during 28 d of storage at 4 °C. Yogurt flow behaviors best fit the Herschel-Bulkley model and showed pseudoplastic behavior. Flow behavior index and yield stress values of yogurt did not change significantly during storage. Furthermore, yield stress was not affected by fortification. On the other hand, yogurt firmness increased during storage, and yogurts fortified with higher levels of calcium phosphate and zinc oxide nanoparticles had greater firmness compared to unfortified yogurts. Santillan-Urquiza et al. (2017) attributed this greater firmness to binding of zinc and colloidal phosphate to the casein micelles.

5 Rheological Changes in Yogurt Resulting from Processing, Storage and Handling

Although many aspects of processing play important roles in yogurt body and texture, temperature has the greatest impact, beginning with yogurt base processing temperature and continuing through fermentation, cooling, transportation and subsequent storage, and even the mastication process. The following three subsections focus on recent research in yogurt processing, fermentation, storage, and handling conditions that impact rheological properties of yogurt.

5.1 Yogurt Base Processing

It is well known that heating milk above 70 °C causes denaturation of whey proteins and promotes interaction of those denatured proteins with caseins. When combined with acidification, either direct or from lactose fermentation by lactic acid bacteria, a gel network is formed. Compared to acid gels made from unheated milk, acid gels from heated milk have more solid-like behavior, reportedly because of increases in heat-induced interactions between caseins and whey proteins that form a stronger gel (Lucey and Singh 1997). For semisolid foods like yogurt, there is a close relationship between their rheological properties and the degree of protein denaturation during the heating process. Heating time and intensity determine the amount of α-la and β-lg bound to casein. It has been reported that under lower heat treatments, filaments of denatured β-lg on the casein micelle surface prevent micellar fusion, while at higher intensity treatments, α-lac segments precipitate onto the micelle, leading to smoother micellar surfaces and improved rheological properties (Mottar et al. 1989; Benezech and Maingonnat 1994).

It is believed that in fortified yogurts that are typically subjected to high heat treatments, coagulation happens in two stages: one at pH 5.3, the isoelectric point of β-lg, and the second at pH 4.6, the isoelectric point of casein. It is desirable that the time between these two coagulations be short, as the lower extent of interaction between the casein micelles leads to a smoother texture. Single-culture strains have longer fermentation times compared to combinations of starter cultures, and this longer time can negatively impact the smooth texture and viscosity of the fortified yogurt (Sodini et al. 2002).

Ozcan et al. (2015) prepared yogurt gels using commercial starter cultures after reconstituting skim milk and adjusting to pH 6.2, 6.7, or 7.2. After heating at 85 °C for 30 min, a portion of the heated milk samples was readjusted to pH 6.7; all samples were inoculated with 3% (w/w) yogurt starter culture and incubated at 40 °C to pH 4.6. Storage moduli values at pH 4.6 were highest in gels made from milk heated at pH 6.7 and lowest in milk heated at pH 6.2, with or without pH adjustment after heating; G' values at pH 4.6 were lower in samples after adjustment back to pH 6.7 after heating. Interestingly, microstructural differences were not observed among the treatments. The authors concluded that heating milk at its natural pH (~6.7) created an optimum balance of casein-bound and soluble denatured whey proteins, which resulted in yogurt with the highest gel firmness.

Riener et al. (2010) used thermosonication to investigate the impact of combining homogenization and pasteurization into a single unit operation on yogurt characteristics. Preheated (45 °C) milk with varying levels of fat (0.1%, 1.5% and 3.5%) was thermosonicated for 10 min at an ultrasound frequency of 24 kHz (400 W) and compared to control yogurts produced from conventionally heated milk (90 °C for 10 min). Yogurts from the thermosonicated milks had higher firmness, higher water-holding capacity, and lower syneresis. Preference tests (n = 30 consumers) revealed that panelists preferred the texture of the thermosonicated yogurts, indicating promise for the technology.

To more closely examine the impact of ultrasound on gel formation, Madadlou et al. (2010) conducted dual-frequency sonication on casein solutions (3% casein in 0.5 M phosphate buffer) to acidification. Model casein gels were prepared from solutions sonicated with 24 (low frequency or power ultrasound) and 130 kHz (medium frequency or sonochemical ultrasound) for 0, 60, or 120 min, followed by acidification with GDL (0.23 g GDL/g casein) at 30 °C. Sonication of casein solutions increased gelation time, postponing the gelation point to a lower pH, and increased the firmness and solid-like behavior of freshly-formed gels. Microstructural images revealed gels made with dual-frequency sonication had more interconnected microstructures and smaller non-distinguishable particulates, particularly for the gel made from the solution sonicated for 120 min. Madadlou et al. (2010) concluded that dual-frequency sonication may be an option for increasing the firmness of fat-free and low-fat yogurts that suffer from weak body and poor texture.

5.2 Yogurt Fermentation

Standard conditions for yogurt fermentation are meant to promote the metabolism of *S. thermophilus* and *L. bulgaricus*, which thrive at ~42 °C and 37 °C, respectively. The most vital part of yogurt production is the fermentation of lactose to lactic acid by the starter cultures. This production of acid decreases the yogurt mix pH and hence forms the gel. During fermentation, flavor compounds are produced that impart characteristic flavor to yogurt, such as acetaldehyde and lactic acid. The final characteristics of yogurt depend on the various factors during the fermentation process, which include fermentation temperature, starter culture selection (e.g. species and subspecies), and yogurt mix composition and treatment.

Lee and Lucey (2006) studied structure–function relationships between the initial yogurt gels and stirred yogurts made from these gels. Yogurt gels were made from milk preheated at 75 or 85 °C for 30 min, inoculated at 2%, and incubated at 32, 38, or 44 °C; then the gels were sheared at 5 s^{-1} for 1 min to make stirred yogurts. Gelation time decreased and pH at gelation increased when heating and incubation temperatures were increased. Set yogurt preheated at 85 °C had branched and cross-linked microstructures, while those preheated at 75 °C exhibited thinner strands and clusters in the protein network. Furthermore, the yogurts with lower heat treatment during pasteurization had higher oral viscosity and lower chalkiness in sensory tests (n = 10 trained panelists). Set yogurts incubated at 32 °C had more interconnected protein structure than the set yogurts incubated at 38 or 44 °C. Stirred yogurts exhibited much more dense protein aggregates, likely because stirring destroyed the initial network, yielding subsequent formation of weak aggregates. Tighter, interconnected structures were associated with higher firmness. Lee and Lucey (2006) concluded that the structure of the initial gel network as well as the structural breakdown process had a major impact on the physical and sensory attributes of stirred yogurts: initially weak protein networks produced weak stirred yogurts. Higher preheating temperature and lower incubation temperature resulted in higher values of apparent viscosity, G', oral viscosity, and sensory mouthcoating and smoothness for both set and stirred yogurts.

Because milk pH influences the amount of casein-bound, insoluble colloidal calcium phosphate (CCP), Peng et al. (2009) hypothesized that varying fermentation time would influence the rate and extent of solubilization of CCP during any subsequent gelation process. Yogurt base milk pH was varied to pH values ranging from 6.55 to 5.65 by pre-acidification with GDL for 4 hr at 40 °C. The fermentation time, or time to reach pH 4.6 from the initial pH, was also varied from 250 to 500 min by adding various amounts of culture at 40 °C. Pre-acidification increased the solubilization of CCP, increased the early loss of CCP crosslinks, and produced weak gels. Longer fermentation times resulted in greater loss of CCP at the pH of gelation, increased the possibility of greater casein rearrangements, and likely contributed to the increase in whey separation. Fluorescence micrographs revealed that the yogurt gels made with low pre-acidification pH values or long fermentation times had larger clusters and fewer interconnections; gels tended to be weak and had higher whey separation. On the other hand, higher pre-acidification pH values or short fermentation times yielded gels with more branching, greater interconnectivity, and a finer network structure. From these results, Peng et al. (2009) noted that pre-acidification was not recommended for the yogurt industry.

5.3 Yogurt Storage, Handling, and Sensory Evaluation

Over the years, in an effort to increase shelf-life, the viability of yogurt bacteria during storage has been studied. However, few published studies have focused on the impact of storage on the rheological and textural properties of yogurt. In yogurt, pH decreases during refrigerated storage because of the residual metabolic activity of the starter cultures (Marafon et al. 2011). This change in pH can negatively impact the structure of the yogurt gel, leading to breakdown of yogurt structure and typically an increase in syneresis.

Surprisingly, little post-acidification was noted in set-type yogurts made with whole and skim milk stored at 10 °C for 91 d (Salvador and Fiszman 2004). In addition, Salvador and Fiszman (2004) investigated the sensory, biochemical, and textural changes during accelerated (20 and 30 °C) and refrigerated (10 °C) storage of set-style yogurts compared to fresh samples. Syneresis was evident after 1 day of storage, particularly for yogurts stored at 30 °C, which exhibited the most syneresis throughout storage. Firmness increased significantly with storage at all three temperatures, and nonfat yogurt had higher firmness values. Sensory analysis on yogurts at 10 °C revealed that most changes occurred in the first week of storage; subsequent changes were less noticeable. Nonfat yogurt samples that were stored for long times had highest syneresis, and sensory firmness, maintenance of shape, chalky mouthfeel. Higher firmness, astringency, and chalky mouthfeel were associated with lower consumer acceptability scores. Salvador and Fiszman (2004) concluded that data collected at the three storage temperatures could serve as good predictors for physical characteristics of yogurt.

Instrumental analyses are more meaningful if they relate to consumer acceptability of products, so ensuring that instrumental and sensory measures align is a major area of focus in the literature. Harte et al. (2007) found that yield stress significantly correlated ($p < 0.001$) with sensory initial firmness perceived by trained panelists in both laboratory-made and retail yogurts. Apparent residual stress was significantly correlated with sensory viscosity for retail yogurts. Yogurt yield stress had more power than apparent residual stress to detect differences in initial firmness. Thus, not only were fewer samples required for evaluation, but yield stress was considered a good predictor of the sensory initial firmness perceived by panelists. Harte et al. (2007) noted that the use of yield stress as a sensory predictor could reduce the need for training panelists and conducting sensory panels and could offer the potential to manufacture yogurts with targeted yield stress and viscosity properties.

Tribology, the study of friction, lubrication, and wear, has recently emerged as an extension of rheology. It is a method that has been used to explain the lubrication behavior between oral surfaces while eating a food (Prakash et al. 2013; Sonne et al. 2014). In a typical friction test, a stainless steel ball represents the palate and a elastomer pad, such as styrene butadiene rubber, with a regularly structured surface simulates the roughness, softness, and deformability of a human tongue (Sonne et al. 2014). Sonne et al. (2014) evaluated the effects of fat, protein, and casein to whey protein ratio on the lubricating behaviors of stirred yogurt and related those behaviors to sensory properties, including graininess, viscosity, and creaminess. A decrease in friction (and sensory graininess) was associated with decreased proportion of whey protein and increased fat and protein level. These yogurts were also perceived as creamier. Sonne et al. (2014) noted that because of the complexity of the eating experience, the predictive ability of in-mouth viscosity and in-mouth creaminess was improved by combined assessments of rheological, particle size, and tribological characteristics compared to each individual instrumental measurement. Further, they suggested that greater understanding of the key drivers for creaminess would allow food manufacturers to develop reduced-fat dairy products without compromising sensory properties.

More recently, Nguyen et al. (2017) evaluated the effect of different hydrocolloids on texture, syneresis, rheology, tribology, and sensory texture and mouthfeel of set yogurts. Gelatin (0.5–1.5%), xanthan gum (0.005–0.015%), carrageenan (0.01–0.08%), and modified starch (0.5–1.5%) were incorporated into yogurts with 0.1, 1.3, and 3.8% fat. In general dispersion of fat particles within the protein network reduced gel strength. Full fat yogurt had lower viscoelastic moduli values than nonfat yogurt, and less syneresis and increased lubrication ability than reduced-fat and nonfat yogurts. Addition of gelatin to the yogurt formulations reduced syneresis and increased viscosity, gel strength and lubrication properties of the nonfat yogurt. These yogurts also had the same sensory scores for thickness, smoothness and creaminess as full-fat yogurt. Both xanthan gum and carrageenan increased the firmness and viscosity of nonfat yogurt but also significantly increased syneresis and chalkiness and lumpiness attributes. On the other hand, modified starch slightly improved the lubrication properties and sensory thickness of nonfat yogurt without significant changes in chalkiness or lumpiness.

6 Opportunities for Future Research

Yogurt is, and will likely continue to be, one of the most beloved dairy products in the world, in large part because of the multitude of styles, flavors, and rheological properties available to consumers. Moreover, as consumers are becoming more health conscious, consumption of low-fat, probiotic, and prebiotic yogurts is expected to continue and potentially expand. Although a great deal of literature is available about yogurt rheological properties and their ability to indicate sensory textures, some gaps remain and are worthy of research attention:

1. Most literature studies have investigated rheological properties during a relatively short shelf life, up to 30 d. Since commercial shelf life could be from 60 d to perhaps up to 120 d, depending on the product and temperature of storage, the impact of longer storage on rheological behavior of semisolid foods needs to be investigated. Shelf-stable yogurt, which is common in China and other populous countries, was not covered in this chapter. However, with shelf life of these prod-

ucts extending up to a year, changes in the rheological properties of shelf-stable yogurt are worthy of investigation.

2. Currently, there is a lack of robust methods to characterize the structure and rheology of semisolid foods in the primary package. The ability to characterize fundamental rheological behaviors of yogurt without disturbing its structure would be convenient in quality assurance, particularly when comparing results among different laboratories. Development of non-destructive methods will be helpful to characterize the effect of formulation, processing and storage conditions, as well as package shape and size, on the properties of the product during storage.
3. There is little information connecting the rheological behavior of semisolid foods to their micro- and nano-scale structural elements using advanced microscopic and spectroscopic techniques. Such information would be helpful in creating a fundamental understanding of rheology and building mechanistic models of rheological changes during storage.

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