



Multiscale Approach to Dairy Products Design

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Dairy products are among the most popular nutritious foods in the world. Understanding the relationship between the composition, process, and structural properties at different scales (molecular, microscopic, and macroscopic) is fundamental to designing dairy products. This review highlights the need to analyze this relationship from different scales as an essential step during product design through a multiscale approach.

Keywords: multiscale, dairy, product design, molecular properties, microscopic properties, macroscopic properties

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1 INTRODUCTION

Dairy products are foods derived from milk that provide nutrients such as proteins, fatty acids, vitamins, and minerals, essentials for function, growth, and the human body's development (Khan et al., 2019; Tunick and Van Hekken 2014). The high nutritional consumption of dairy products worldwide has resulted in the development of novel techno-functional products in the dairy industry (Farhana et al., 2018). Unfortunately, the design of foods using novel ingredients and technologies has been shown to affect the sensory profile and food structure, which consumers perceive as texturally less attractive (Aguilera 2006; Mousavi et al., 2019).

The structure of dairy products consists mainly of milk proteins, fat, water, lactose, and salts. After processing, these molecular components experience changes that define dairy food's typical appearance, taste, aroma, and texture (Spyropoulos et al., 2019). Generally, the food industry monitors its products by controlling their properties during the production and formulation stages. Consequently, the molecular interactions between the components, ingredients, and additives that are part of the formation and structuring are usually not considered during the food production process. Therefore, understanding the relationship between dairy products and their structure is essential to designing novel products that can be more attractive to consumers (Dickinson 2014; Heertje 2014).

Food structure can be considered the link between molecular, microscopic, and macroscopic properties. At the molecular scale, the complex interactions determine the subsequent microscopic properties. At the microscopic scale, it is possible to decide on the textural and rheological properties at the macroscopic scale (Crilly et al., 2008). Understanding the relationship between composition and formulation, the manufacturing process, and the properties of food through these scales is therefore essential for designing a product with specific sensory, stability, shape, and nutritional properties (Ho et al., 2013).

A way of understanding, predicting, and studying the relationships between scales during product design is using a multiscale approach (Crilly et al., 2008; Ho et al., 2013; Huc et al., 2016). The multiscale approach creates a geometric model of the food structure and correlates the scales. This approach allows the analysis and study of behaviors associated with food development's process, product, and properties (Ho et al., 2013). It allows taking advantage of molecular interaction on microscopic and macroscopic properties to design a food structurally.

Consequently, the multiscale approach can be considered an essential tool for designing new dairy products.

Recent research focuses on understanding food structure without jointly considering texture, rheology, microstructure, and the behavior of molecular components involved in food structure formation. This review emphasizes the relevance of considering the interactions between the molecular, micro, and macro scales and their relationship with the steps of the dairy product development process. It provides a comprehensive overview of the factors that affect the structure of dairy products and their relationship with other food properties. This review's scope considers the following dairy products: yogurt, cheese, ice cream, and butter.

2 DAIRY PRODUCT DESIGN FROM A MULTI-SCALE PERSPECTIVE

Dairy products are foods mainly associated with the human body's development and functionality (Pereira 2014). These products are either high milk protein (e.g., yogurt and cheese) or high-fat content (e.g., butter and ice cream). Yogurt is produced by the fermentation of milk with lactic acid bacteria, while cheese is made by adding rennet enzyme (Chymosin and pepsin) to the milk (Guinee 2016). When milk proteins (casein)

coagulate in yogurt and cheese, they form three-dimensional protein networks that trap fat (El-Bakry and Sheehan 2014; Giha et al., 2021; Guinee 2016). On the other hand, butter is a milk fat-based product generally characterized as a water-in-oil emulsion. (Panchal et al., 2021; Truong et al., 2016). Ice cream is a complex multiphase structure containing protein, water, crystallized fat, air, minerals, additives, and sweeteners (Kaleda et al., 2018).

The structure of dairy products can be analyzed through properties at the molecular, microscopic, and macroscopic scales (Figure 1). Mixtures of molecular components such as proteins, fat, water, carbohydrates, salts, and the interactions between water-casein, casein-casein, and casein-minerals define the dairy product structure (Heertje 2014). While these molecular elements can be studied considering pH changes, ionic strength, or hydrophobic interaction (Lucey and Horne 2018), the microscopic scale shows the spatial distribution and particle size/shape of these components, and the macroscopic scale reflects the perception of them in textural and rheology properties (Figure 1).

The formation of the structure of a food is equally dependent on processing (Dickinson 2014). The manufacturing process, like heating, gelation, fermentation, and ripening, among others (Smith et al., 2017), significantly affects the structure of molecular components, which in turn generates changes in the dairy food structure (Joardder et al., 2015). Therefore, ingredient

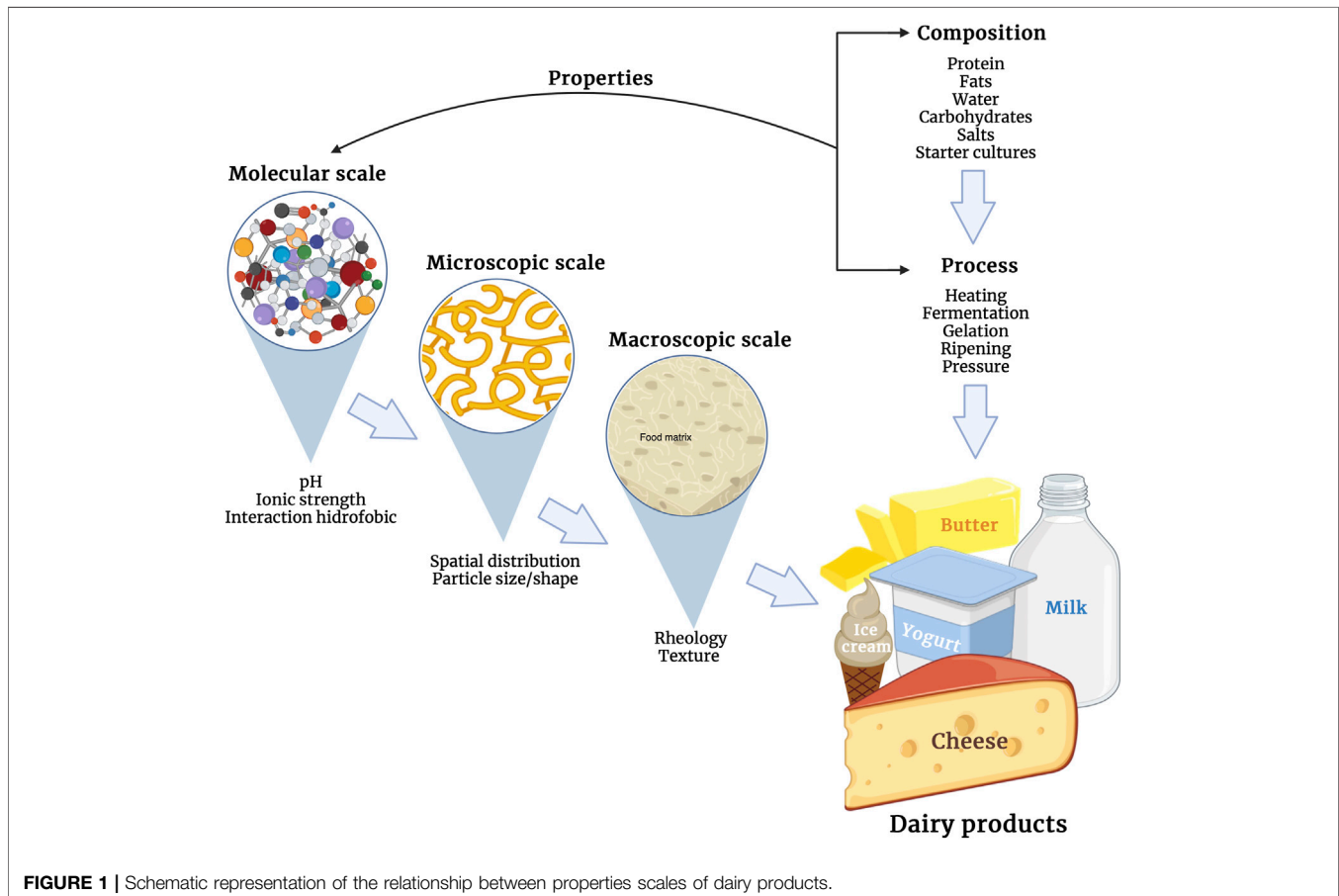


FIGURE 1 | Schematic representation of the relationship between properties scales of dairy products.

composition and processing conditions are mechanistically related to product properties (Aguilera 2006; Dickinson 2014) (Figure 1).

Few studies about dairy products have integrated the relationship between product-process-properties from the multi-scale perspective. For example, Huc et al. (2016) investigated different yogurts' physical properties, analyzing the correlation between fat and protein content, size particle, and rheology properties under temperature conditions, considering the tribology as element additional. As a result, the authors showed that it was possible to obtain information relevant to yogurts' formulation to improve eating. As another example, Díaz-Bustamante et al. (2020) studied the substitution and reduction of NaCl in Costeño-type artisan cheese applying multiscale modeling. This study showed that it was possible to obtain cheeses with physicochemical composition and textural properties similarities of a cheese control, making variations in the NaCl addition and the manufacturing process. The previous examples show that the dairy products design's main challenge is understanding the association between structural scales, composition, and processing, determining the final structure. Therefore, one way to approach this is to visualize dairy products as foods with a multiscale structure where fundamental molecular, microscopic, and macroscopic properties (contributing to structure and sensory characteristics) can be addressed.

2.1 Dairy Products on a Molecular Scale

Food structure is primarily based on their molecular composition, so small changes at this level can induce dramatic structural changes (Barroso da Silva et al., 2020; Heertje 2014). Caseins, fat, lactose, and salts define the molecular composition of a dairy product. Milk is a colloidal dispersion of these molecular components (Foegeding 2015). These components play a significant role in dairy products' stability and nutritional value and contribute to their micro and macroscopic properties.

Caseins are the main molecular components defining the structure of dairy products (Spyropoulos et al., 2019). Thus, understanding casein's network is crucial for improving its processing stability. Caseins are phosphorylated and hydrophobic proteins that can self-assemble and interact strongly with multivalent cations involving molecular properties like ionic strength, hydrophobic interaction, and pH (Lamichhane et al., 2018; Spyropoulos et al., 2019). These proteins are classified into four groups (i.e., α_1 -, α_2 -, β -, and κ -caseins) representing 80% of total milk protein. During the cheese and yogurt manufacturing process, κ -casein, responsible for the electrostatic and steric stabilization of casein micelles, is destabilized by rennet enzymes leading to fermentation or gelation. As a result, it forms a three-dimensional network of protein aggregates, whose stability depends on temperature and mechanical action (Loveday et al., 2013; Moussier et al., 2018). These processes change the residual charges on the casein molecules and the electrostatic and hydrophobic interaction strength between them (Spyropoulos et al., 2019).

Dairy from cows may be subjected to thermal treatments to reduce the growth of pathogenic bacteria (Wiking et al., 2021).

However, heating milk to over 70°C leads to the denaturation of whey proteins, resulting in protein complexes formed from disulfide bonds with κ -casein, and this, in turn, affects manufacturing processes such as cheese coagulation (Asaduzzaman et al., 2021; Giroux et al., 2020; Lucey et al., 2021). It is worth noting that not only does heat affect the structure, conformation, and properties of whey proteins, but also these proteins are affected by pH and ionic strength (Anema 2019; Ong et al., 2020). Although milk is treated with heat in the manufacture of cultured dairy products to increase viscosity and improve texture (Lucey et al., 2021; Zhao et al., 2016), this process also induces several molecular changes in the milk fat globule affecting its functionality (Wiking et al., 2021).

The pH is another parameter that influences the fermentation and gelation of caseins and modifies their net charge (Ong et al., 2012). In the case of milk, the pH value will vary depending on the effect of the starter cultures or acidulant added (Ong et al., 2012; Spyropoulos et al., 2019). For instance, during fermentation of yogurt, lactic acid bacteria reduce the milk pH to the isoelectric point of casein, which is pH 4.6, leading to micelle aggregation because of the reduction of the net negative charge of proteins (Zhao et al., 2016). In cheese, milk pH affects the way casein micelles aggregate. At lower pH, there is an increase in the number of Ca^{+2} ions in solution, which reduces the surface potential of the κ -casein and causes dissociation between casein micelles (Ong et al., 2012). On the other hand, pH can be considered a molecular property and a process variable that can increase yield and alter texture in Cheddar cheese (Ong et al., 2012).

Interactions of caseins with other molecular compounds, such as fat globules, water, and minerals, also play an essential role in defining the structure of dairy products. For instance, the fat globule size can alter the casein strand formation during rennet-induced coagulation of milk (Lamichhane et al., 2018). On the other hand, the function of fat globules in ice cream is different; they have a specific role in stabilizing the bubbles and are responsible for the product's creamy texture (Bhopatkar et al., 2012). The interactions of water-casein through hydrogen bonding can increase the moisture content and be measured as the water capacity of a protein or the total amount of water that can be absorbed per protein weight (Aryee et al., 2018; Lamichhane et al., 2018). The addition of calcium chloride influences the stabilization of casein micelles, and it is commonly used to improve clot formation and increase cheese yield (Soodam et al., 2015). The addition or reduction of salts such as NaCl or KCl can also affect the stability of casein, which influences textural and rheological properties in products such as Costeño-type artisan cheese (Díaz-Bustamante et al., 2020).

All the above-mentioned molecular components and their structure differ among dairy products after processing; as a result, these various structures also impact their micro and macrostructure properties (Aguilera 2006). Therefore, this correlation can be analyzed by scanning electron microstructure (SEM), rheology behavior, and texture (Huc et al., 2016; Loveday et al., 2013; Verboven et al., 2018). Another experimental method can be zeta potential, which has been assessed under different processing conditions and milk

TABLE 1 | Current strategies used in the development of new dairy products.

Product	Composition	Process stage	Properties			Strategy	Authors
			Molecular	Microscopic	Macroscopic		
Yogurt	—	—	pH	—	Rheology (storage modulus, G') and sensory properties.	Product + properties Product + properties	Pachekreppapol et al. (2021) Vianna et al. (2017)
	—	—	pH and titratable acidity	Scanning electron microscopy	Texture (firmness and apparent viscosity)		
Cheese	Fat and sodium content	Curd washing, moderate clotting, scalding, temperature, ripening, and high pasteurization	—	—	Texture (hardness, cohesiveness, adhesiveness, and gumminess)	Process + properties	Moatsou et al. (2019)
	Microparticulated whey protein	—	—	—	Rheology (storage modulus, G'), texture (firmness, adhesiveness and cohesiveness), and Sensory evaluation	Product + properties	Stankey et al. (2017)
Ice cream	—	High-pressure homogenization and melting	—	Scanning electron microscopy	Texture (hardness, adhesiveness, gumminess, and cohesiveness)	Process + properties	Sert et al. (2021)
	—	Fermentation	—	Scanning electron microscopy	Rheology (apparent viscosity, consistency coefficient, and flow behavior index)	Process + properties	Dertli et al. (2016)
Butter	—	Ripening and churning	—	—	Rheology (elastic modulus and viscous modulus)	Process + properties	Mtibaa et al. (2021)
	—	High-pressure homogenization	—	—	Texture (firmness, consistency, cohesiveness, and viscosity index), spreadability, and particle size	Process + properties	Sert and Mercan (2020)

treatments for analyzing the stability of casein micelles and milk fat (Cano-Sarmiento et al., 2018).

2.2 Dairy Products on a Microscopic Scale

Microstructural characterization is critical for understanding food properties and functionality (Auty 2011). Knowledge of food's microstructure is important for designing and controlling food processing operations and improving the quality of the final product (Joardder et al., 2015). Microscopic properties in food are characteristics, such as air pores, microcapillaries, cells, and fibers that can be found on a length scale of 0.1–100 μm , forming a continuous body that requires light microscopy to be visualized (Auty 2011).

The microstructure is determined by complex molecular interactions and depends mainly on the organization of molecular compounds in the food matrix (Verboven et al., 2018). In dairy products, the microstructure is based on the spatial distribution of caseins, fat globules, water droplets, and air spaces (Fox et al., 2016). Protein and fat are the prominent influencers of the microscopic properties of food (Auty 2011; Truong et al., 2016; El-Bakry et al., 2018). However, in dairy products such as ice cream, the microscopic properties depend on the fat and protein content and the whey phase, ice crystals, and air (Goff and Hartel 2013). Electron microscopy is one way to understand the structure of foods. In the case of dairy products, electron microscopy makes it possible to observe casein micelles and their interactions with other components (Ong et al., 2013; El-Bakry et al., 2018).

Caseins are approximately 130–160 nm and can form microstructures in a wide density range depending on manufacturing conditions like heat treatment (Díaz-Bustamante et al., 2020; Lucey et al., 2021; Spyropoulos et al., 2019). For instance, acid-induced dairy gels from heated milk are less porous than unheated milk and exhibit a homogeneous and rigid microstructure (Guinee 2016; Spyropoulos et al., 2019). Moreover, high processing temperatures produce a less dense crystal network and a protein/water distribution change in butter (Buldo and Wiking 2012).

The fat structure also plays a crucial role in the microstructure (Amador et al., 2017). It is related directly to sensory properties like mouthfeel, creaminess, flavor, and product stability (Crilly et al., 2008). Fat globules have a size of around 0.1–15 μm (Lamichhane et al., 2018; Wiking et al., 2021) and can be seen in different morphologies in the microstructure of dairy products; for example, in the form of round droplets in the milk, like crystals in ice cream, or as a kind of amorphous phase in cheese (El-Bakry et al., 2018). Studies have shown that fat globule (FG) affects the physicochemical composition and macroscopic properties such as firmness and hardness (Logan et al., 2017; El-Bakry et al., 2018; Panchal et al., 2021). For instance, Logan et al. (2017) indicated that the FG size increased the moisture content in Cheddar cheese affecting its hardness.

In high-protein products, such as cheese, fat globules are usually trapped in the protein network, preventing their movement, and providing a firm texture to the product. However, during the ripening process, proteolytic enzymes

alter the protein network, allowing the fat globules to escape, producing the characteristic oily appearance of aged cheese (Bhopatkar et al., 2012).

In recent studies, dairy products have been analyzed both by the association between fat and proteins (Macdougall et al., 2019) through different micrographic analysis techniques such as light microscopy, confocal scanning laser microscopy, scanning electron microscopy, and transmission electron microscopy (El-Bakry et al., 2018). However, few studies determine the microscopic properties through a global analysis of the influence of physicochemical composition, process conditions, and how the behavior and functionality of the molecular components are reflected in the microstructure of the dairy product.

2.3 Dairy Products on a Macroscopic Scale

Macroscopic properties can be defined as the spatial distribution of microscopic and molecular properties (Foegeding 2015). Such properties are associated with tangible and visible properties of the food, which can be measured by the naked eye (Ho et al., 2013).

Rheology and texture are macroscopic properties that describe the association of physical properties with mouthfeel and sensorial perception of food (Nguyen et al., 2018). Rheology is the science of deformation and flow, aimed at linking a material's flow properties with its structure and behavior during processing (Truong et al., 2016); These properties are measured through flow sweep, flow ramp, and oscillatory measurements. Textures are all mechanical, geometric, and surface attributes of a product perceived mechanically and tactilely (Pascua et al., 2013). The sensory attributes describe by textural analysis in foods are hardness, springiness, cohesiveness, adhesiveness, gumminess, and chewiness.

Rheological parameters and texture, as well as microscopic properties, depends on the interaction between proteins (Heertje 2014), pH (Ozcan et al., 2015), state of calcium (ionic or bound to the casein matrix) (Soodam et al., 2015), sodium chloride content (Díaz-Bustamante et al., 2020) and temperature (Asaduzzaman et al., 2021). However, these components can govern the different macroscopic properties of dairy products. For instance, Ong et al. (2020) studied the effect of pH variation on cream cheese production. These authors indicated that the microstructure was dense and cheese texture firmer as the pH decreased. In addition, other components also can govern the different macroscopic properties of dairy products. The rheological properties of ice cream are governed by ice crystals (Freire et al., 2020), whereas in butter, these properties are controlled by the crystalline structure of the fat (Buldo and Wiking 2012; Rønholt et al., 2013).

Some studies show that molecular components like fats and calcium content can affect dairy products' rheological properties and texture (Soodam et al., 2015). Macdougall et al. (2019) indicate that the hardness and gel strength generally correlated with high fat and high content increase the overall cohesiveness of Australian cream cheese.

Concerning process variables, heat treatment also affects these macroscopic properties (Buldo and Wiking 2012; Zhao et al.,

2016; Díaz-Bustamante et al., 2020). Denaturation of milk proteins by heating and interaction among denatured milk proteins dramatically affects the texture attributes, consistency, and storage moduli of dairy products (Zhao et al., 2016; Giroux et al., 2020; Lucey et al., 2021).

3 LOOKING FORWARD

The development of novel and functional dairy products has been of great interest to the dairy industry in recent years (Guiné and Lemos 2018). The demand for products with high nutritional value, with the incorporation of bioactive compounds, reduction or addition of fats, salts, and sugars with more significant contribution to health, has become a challenge for the design of these products because these ingredients or substitutes can modify the structure of dairy products (Meyer et al., 2011; Mudgil and Barak 2017; McCain et al., 2018). For example, inulin is a hydrocolloid that is considered a functional ingredient used as a fat substitute in dairy products (Li et al., 2019; Yousefi and Jafari 2019); however, Meyer et al. (2011) indicate that its incorporation is related to changes in the rheology of the foods. Yousefi and Jafari (2019) reviewed the functionality and application of various hydrocolloids in dairy products, including inulin. These authors noted that hydrocolloids affect dairy products' physical properties. These changes depend on the interaction with protein, pH, temperature, ionic strength, chemical structure, and the concentration of hydrocolloids.

Many of the advances in the structural development of dairy products have focused on the application of new technologies such as ultra-pressure (Ravash et al., 2020), high-intensity ultrasound (Guimarães et al., 2019), microfluidization (Bucci et al., 2018), or pulsed electric fields to improve the texture of foods (Sfakianakis and Tzia 2014). The purpose of this is to improve the properties of the proteins with minimal changes in their organoleptic and nutritional characteristics (Ravash et al., 2020). For example, Siddique et al. (2016) indicate that pulsed light treatment improves whey protein's solubility and foaming ability. However, Nunes and Tavares (2019) highlight that these emerging technologies can modify the milk proteins' structure and techno-functional properties.

Other studies have pointed out the importance of microstructure, rheology, and textural attributes of dairy products, including manufacturing conditions and changes at molecular scale during the development of the food (Dertli et al., 2016; Huc et al., 2016); however, the analysis of all these elements as a whole remains limited. For instance, Pachekrepapol et al. (2021) developed a yogurt formulation from coconut milk. In this study, the authors evaluated the pH changes during storage (molecular scale) and the product's rheological and sensory properties (macroscopic scale), considering syneresis, color, and heating process. However, the authors have forgotten the effects caused by the incorporation of coconut milk on the microstructural properties of yogurt. Unlike the multiscale approach, similar studies have developed new dairy products through strategies that do not include the relationship between product-process-properties. A summary of these studies is shown in **Table 1**.

A better understanding of their properties can address the design of dairy products at different scales, as well as the interconnection between them (i.e., a multiscale approach), which could lead to the estimation of structural properties, optimize the process, and develop new foods (Ho et al., 2013). The multiscale approach allows understanding the relationship between composition and formulation, the manufacturing process, and the properties of food through properties at the molecular, microscopic, and macroscopic scales, allowing the development of foods with specific desired properties.

Dairy products design at the molecular scale allows knowing all the interactions that occur in molecular compounds once they are subjected to process conditions and how these are associated with the formation of the product's structure. Then, dairy products design at the microscopic scale shows the spatial distribution of the molecular components and how this consequently affects the structure of the food. Finally, dairy products design at the macroscopic scale predicts the final characteristics through rheological and textural responses associated with tangible and visible properties of the food.

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