7 Design Structures for Controlled Manipulation of Flavour and Texture

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7.1 NEED FOR CONTROLLED FLAVOUR AND TEXTURE FOOD DESIGN

Many foods are multiphase systems with complex structures, which to a large extent determine the sensorial (e.g. taste, aroma, texture) and nutritional (e.g. digestibility, physical, chemical and microbial stability, and bioavailability of nutrients) properties of the food. The role of microstructure in food functionality and customer appeal is often linked with length scales of 0.1–100 μm, as many of its structural elements (such as starch granules, protein assemblies, polymer networks, solid crystals, small particles, oil droplets and gas bubbles) fall within this scale range (Aguilera, 2005). Interestingly, there is a recent trend in food research to extend the range of interest towards smaller entities, and to study the role of nano-structures in determining and manipulating the characteristics and quality of food products (Aguilera, 2005).

Understanding and quantifying the nature of the relationship between the structure of a food and its properties is far from straightforward, and poses a fascinating academic challenge, conveniently combined with an attractive potential for the food industry. Creaminess, for example, is an extensively investigated microstructure-dependent food quality (Kilcast and Clegg, 2002), yet it still remains a challenging research topic showing product specific characteristics (Mosca *et al*., 2012; Yamul *et al*., 2013). In the case of ice cream, essentially a multiphase system consisting of air, crystalised fat, and ice suspended in a concentrated sugar solution containing hydrocolloids and proteins, microstructure determines a number of sensorial features, such as gumminess, crumbliness, coarseness, smoothness, sogginess, and percep-

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tion of optimal creaminess has been linked with stable, small-sized air bubbles (Eisner *et al*., 2005). It comes to no surprise, therefore, that the industry has concentrated efforts on producing ice creams with stable, optimal microstructures.

Structured foods also have great potential to address specific diet-related diseases, such as undernourishment, allergies and obesity, which were characterised as a "modern epidemic" 25 years ago (Seely *et al*., 1985), and are ever increasing. Encapsulation technology is often engaged to formulate foods enriched in vitamins, minerals or bioactive compounds (Kaufmann and Palzer, 2011). By carefully selecting the encapsulating material, increased bioavailability of the active compound can be attained, resulting from technologies including homogeneous dispersion, protection from the environment and controlled release (McClements *et al*., 2007; Kuang *et al*., 2010). Undernourished populations, the number of which is constantly increasing (Chopra and Sanders, 2008), or individuals requiring supplementation of nutrients, would clearly benefit from such food formulations. Similarly, "free-from" foods, where an ingredient is removed from the food, are designed for people with allergies and/or intolerances. As such, dairy-free yoghurts and cheese equivalents, for example, are well appreciated by individuals with lactose intolerance.

Reducing the contents of specific ingredients (fat, sugar, salt) in foods has been associated with broader health benefits. Notably, energy-rich diets in the USA have resulted in an approximate 200% rise in the reported rate of obesity (a body mass index, mass(kg)/(height(m))^2, greater than 30) in only 25 years time (see Fig. 7.1)! Obesity is known to adversely affect a number of chronic diseases, such as diabetes, asthma, coronary and other cardiovascular diseases, arthritis, osteoporosis and chronic kidney disease (Ross and McGill, 2006; Grande *et al*., 2009). There is thus a currently growing demand for

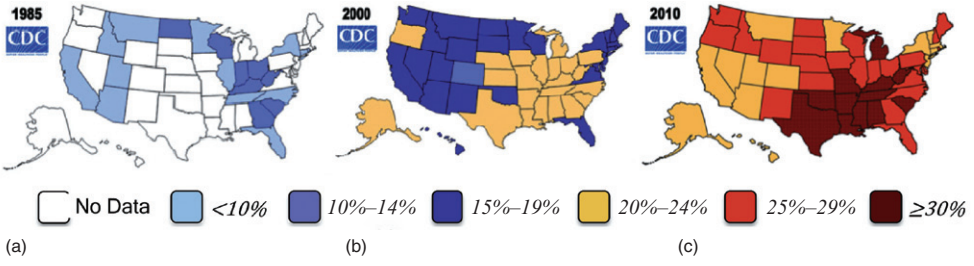


Fig. 7.1 Evolution of reported obesity rate state by state in the USA in (a) 1985, (b) 2000 and (c) 2010. Reproduced from Centers for Disease Control and Prevention (n.d.a) U.S. Obesity Trends. http:/www.cdc.gov/obesity/data/trends.html.

low- energy (low fat and sugar) products, and maintaining the desirable structure without compromising the sensory attributes of the original product becomes increasingly challenging.

Diets high in sodium constitute a serious risk for hypertension, a condition currently affecting a third of the US population (Lindsay *et al*., 1982; Nowson *et al*., 2003). More than 50% of the US population has been reported to consume an average of 3400 mg/day compared to the recommended 2300 mg/day (2008 data), prompting research on strategies to reduce sodium intake (Henney *et al*., 2010). Furthermore, high lipid and cholesterol intake have been associated with risk for coronary heart and Alzheimer's diseases (Petot and Friedland, 2004; Truswell, 2010).

Clearly, the first step to achieving the formation of structures with specific characteristics is an in-depth understanding of the role of each ingredient, and its relationship with the other components of the food matrix. Yet altering the contents or processes of food preparations carries the risk of decline in sensory perception or physical, chemical and microbiological stability (Le Révérend *et al*., 2010; Norton and Norton, 2010). These aspects are typically addressed by identifying and designing the relevant food structures: for example, W/O/W double emulsions have been employed in the formulation of reduced-fat foods (Kaufmann and Palzer, 2011); encapsulated sodium has been demonstrated to increase the sensation of saltiness at reduced salt levels (Morris *et al*., 2009); the addition of hydrocolloids that self-assemble under the stomach's acidic conditions has been linked with increased sensation of satiety (Spyropoulos *et al*., 2011). In this context, the food industry and governmental organisations have recently launched programmes aiming at developing structural solutions to reduce sugar, fat and sodium content in foods (Norton *et al*., 2006).

In order to develop and validate food structures compliant with this design it is important to: (1) understand the conditions that foods are subject to in the mouth; (2) record the sensory sequence that consumers experience, to enable comparison between different engineered formulations and (3) develop a range of *in-vitro* and *in-silico* methods to rapidly prototype candidate structures without the need for repeated, convoluted sensory testing.

As such, this chapter will describe the current understanding of oral processing, and its relationship with sensory perception, and our ability to monitor and model it. Tribology, the measurement of friction and lubrication, will be introduced, as will mouth simulators. Next, the interaction between foods and the oral cavity will be discussed, with reference to the interactions with the saliva and mucus layers overlaying the oral surfaces. Finally, chocolate is given as an example product

to illustrate the interaction between food microstructure and sensory perception.

7.2 ORAL PROCESSING

7.2.1 The oral processing "machinery"

The cavity of the mouth is the entrance of the digestive system, and the region where sensorial attributes of food are mostly appreciated. It broadly consists of two parts: the oral vestibule and the mouth cavity proper. The oral vestibule is the area delimited by the lips, cheeks, gums and teeth, while mouth cavity proper is bounded by the gums and teeth, roofed by the hard and soft palate, and floored by the tongue and sub-tongue mucosa. Saliva is secreted into the mouth cavity proper through the submaxillary and sublingual salivary glands (Gray, 1918). An anatomic diagram of the oral anatomy is shown in Fig. 7.2.

When considering the different properties of the oral cavity that are relevant for in-mouth food manipulation and transformation, one should first consider the actual dimensions of the mouth. Oral dimensions have

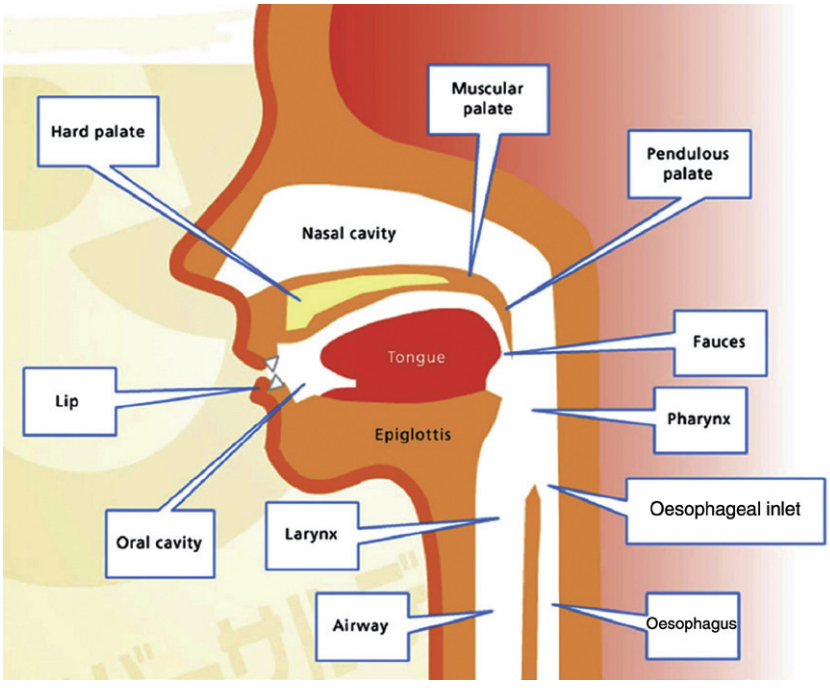


Fig. 7.2 Schematic of oral anatomy. Reprinted from Funami *et al.*, (2012) Texture design for products using food hydrocolloids. *Food Hydrocolloids*, 26, 412–420. Copyright 2012 with permission from Elsevier.

been measured using a range of techniques ranging from calipers (Bakwin and Bakwin, 1936) to 3D laser profilometry scanning (Hohoff *et al*., 2005). Such measures have indicated that a typical adult palate is 34 mm wide, 15 mm high and 40 mm long, which, if assumed parallelepipedic in shape, would represent a volume of 20.4 mL. Interestingly, this number is remarkably close to values reported for a typical mouthful of banana (18 ± 5 g). Dimensions of the oral cavity probably dictate the range of maximum acceptable bite size for optimal manipulation of food.

7.2.2 In-mouth food processing

Typical transformations of food in the mouth occur as a consequence of three main activities: (1) *mastication* and the resulting conversion of consumed food into a swallowable bolus through mechanical breakdown of solids and simultaneous lubrication by saliva; (2) *manipulation* of soft solids (including a masticated food bolus) and liquids between the tongue and hard palate, and their passage towards the pharynx; (3) *swallowing*, which propels foods into the pharynx and oesophagus.

A three-dimensional model to describe food transformations during eating and their link to texture perception has been proposed by Hutchings and Lillford in 1988 (see Fig. 7.3). In their original contribution, the authors represented oral processing as the path of sequential modifications experienced by the food, from entering the mouth (first bite) until swallowing. The model represents changes in the degree of structuring (reduction) and lubrication (increase) of consumed foods as a function of in-mouth processing time. Comparison between five different food systems suggests that the time span of oral processing from the first bite until formation of swallowable material strongly depends on the rate at which structure reduces and lubrication increases for each individual food, as well as on the initial characteristics of the food, predominately its microstructure and physico-chemical properties. This dynamic approach to in-mouth texture analysis, where time forms an integral part of the study, initially saw limited number of followers, which has been at least partially attributed to significant technical challenges and limitations of (or lack of) the relevant quantitative techniques (Chen, 2009). Recently, however, development of time-dependent sensory methods has enabled dynamic measurements and the model has been revisited and strengthened. Findings have confirmed that sensory perception is modified over the course of oral processing in line with the transformations occurring, for solids foods such as breakfast cereal (Lenfant *et al*., 2009) and also in beverages (Le Révérend *et al*., 2008).

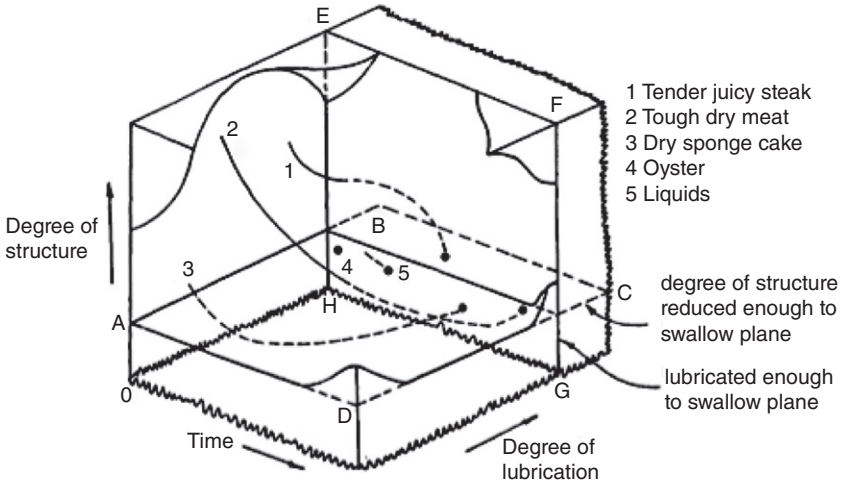


Fig. 7.3 Oral processing model for five different foods. Reproduced with permission from Hutchings and Lillford (1988). The perception of food texture – the philosophy of the food breakdown path. *Journal of Texture Studies* 19, 103–115. Copyright 2007, John Wiley and Sons.

7.2.2.1 *Mastication of solid foods*

Mastication involves chewing of solid foods and mixing with saliva within the oral cavity and has been extensively studied by scientists from the dental, biomechanical and food-science communities. During mastication, solid foods are transformed into swallowable boluses through mechanical disruption (particle size reduction) and lubrication. In this context, a large number of investigations have centred on studying the applied forces, using sensors within the oral cavity (Mioche *et al*., 1993) and consequent reduction in particle size using spit-out experiments (Lucas and Luke, 1983; Van Der Glas *et al*., 1987; Peyron *et al*., 2004). Measurements of particle size distributions in a food bolus suggest particle diameters of around 2–3 mm (Jalabert-Malbos *et al*., 2007).

7.2.2.2 *Manipulation of soft solids*

After initial mastication, or upon consumption of products that do not require mechanical breakdown (e.g. cream cheese or yoghurt), foods are manipulated between the tongue and the palate and are specifically directed for passage from the mouth into the oesophagus and stomach. It is at this stage that many sensory attributes, such as thickness and creaminess, are evaluated. A thorough understanding of the processes occurring during this phase would therefore provide an invaluable tool for the food designer. However, direct (*in vivo*) studies are difficult, chiefly because the relevant physical movements involved are usually

considerably fast and hard to follow. In addition, a non-invasive method would be required, which only adds to the already loaded degree of experimental complexity.

Alternatively, *in vitro* methods can, and have, been used to characterise mixing in the mouth during consumption of, for example, semi-solid foods. Typical mixing times of the order of 10 s were suggested (Prinz *et al*., 2007), although shorter times should be expected for lower viscosity products. Researchers from the same institute have also used ultrasound imaging to evaluate the differences in tongue movements that are carried out during the evaluation of different sensory attributes, such as thick, creamy, sour and bitter (de Wijk *et al*., 2006). They reported that the tongue's movements during intraoral manipulation are mainly influenced by the food's sweetness, whilst food's viscosity mainly influences the swallowing pattern.

7.2.2.3 *Swallowing*

Swallowing of a bolus signals the end of oral processing of a food bite. The processed food is squeezed between the tongue and hard palate (see Fig. 7.2) and further propelled into the pharynx with the aid of a mechanism involving the epiglottis, which ensures that ingested food does not enter the body's airway. The mechanics of this food bolus ingestion have been investigated experimentally using videofluorography (Gates *et al*., 2006), magnetic resonance imaging (Honda and Hata, 2007; Sutton *et al*., 2009) and ultrasound imaging (Stone and Shawker, 1986), the latter offering a non-invasive, safe for subjects method for quantification, at the expense of a somewhat lower image quality. All these measurements have investigated the motion of the soft tissues involved during mastication.

From a food science and engineering perspective, direct experimental measurements of the flow patterns would certainly be more relevant; to the best of our knowledge, this has not been reported yet. Such flow patterns have, however, been investigated numerically by Nicosia and Robbins (2001). Using a squeeze flow analogy, the authors computed the typical Reynolds numbers involved in the swallowing process. Depending on fluid viscosity, they reported Reynolds numbers varying between 10 000 (for fluids with viscosity $\eta = 1$ mPa.s) and 0.1 (for fluids with viscosity $\eta = 10$ Pa.s), although this latter case may not be easily swallowable for a human subject. Such values indicate that flow of a bolus during swallowing can be either turbulent or laminar, depending on the fluid ingested. Knowledge of the flow patterns during swallowing and it is of particular relevance, for example, when designing products for populations with swallowing difficulties (dysphagia).

7.2.3 Sensory feedback during oral processing

Oral processing involves the close interaction between food and the biological tissues of the mouth. The main properties of both interactive parties dictate their inter-relationship and the way food is perceived, which will be briefly discussed in this section.

Taste and texture of foods are sensed in the oral cavity, which in turn feeds the central nervous system (CNS) with sensory information. Textural transformations occurring in the food during oral processing are continuously sensed by the mouth's sensory system. It is this feedback from the CNS that promotes the relevant adjustments in the masticatory parameters for optimal oral processing, and decides when the bolus is suitable and safe for ingestion (van der Bilt *et al*., 2006; Jalabert-Malbos *et al*., 2007).

As food passes over the tongue, it contacts the taste receptors. Receptors for tastes (salty, sweet, bitter, sour, umami) are located on the surface of taste receptor cells (TRCs) that bundle in aggregates to form taste buds. Those taste buds are distributed between the circumvallate, foliate and fungiform papillae of the tongue (Chandrashekar *et al*., 2006). Perception of sourness and saltiness are triggered when the small H^+ and Na^+ ions, respectively, pass through specific ions channels embedded in the lipid bilayer of receptor cells (Mattes, 1997; Chandrashekar *et al*., 2006). By contrast, sweetness, bitterness and umami receptors are triggered by much larger molecular structures than single ions, and are encoded by G-protein-coupled receptors, a process which does not require penetration of the tastant into the TRC. Recent research also seems to indicate that other nutrients may be sensorially encoded by similar mechanisms, such as calcium (Gabriel *et al*., 2009) and fat (Cartoni *et al*., 2010). For more information about this topic, the reader is referred to the review by Chandrashekar *et al*. (2006).

It also seems of relevance that food material scientists should additionally consider the material properties of the biological tissues that are in contact with foods during oral manipulation. Respective values of the Young elasticity modulus (E) for the tongue, soft and hard palate are 10, 25 and 2000 kPa, respectively. As these tissues are mainly composed of water it is not surprising that reported Poisson ratios (ν) are close to 0.5, which is typical of an incompressible material. For the sake of comparison, typical readily swallowable foods, such as apple sauce or fruit purees, have a yield stress in the order of magnitude of 10 Pa (Steffe, 1992), indicating that no major deformation of the tissues occurs during swallowing.

It is most likely that the integrated feedback of those chemical and mechanical sensors constitutes the first chain of command controlling the breakdown pattern described by Hutchings and Lillford (1988).

7.2.4 Monitoring of food oral processing

Monitoring *in situ* phenomena occurring during eating would provide essential information towards understanding phenomena and designing foods. However, as with many processes, such observations pose significant difficulties and experimental challenges. Direct observations of tastants being processed require probing their concentration in the vicinity of the receptors embedded in the TRC membranes, which is very difficult, if not impossible, with the present technologies. In addition, monitoring texture perception is very challenging. As already discussed, food texture is heavily modified during oral processing due to the effects of mastication. Solids experience significant particle size reduction (Lucas *et al*., 2004; Peyron *et al*., 2004; Lenfant *et al*., 2009), while emulsion droplets and foam bubbles are subjected to narrow-gap shears and are therefore prone to coalescence and/or break-up. In addition, lubrication and mixing with saliva transforms the processed food into a coherent bolus ready to swallow.

Besides "chew and spit" experiments and *ex-vivo* rheology (Peyron *et al*., 2004; Lenfant *et al*., 2009), the current state-of-the-art techniques to monitor those changes in real time are still based on sensory analysis. In recent years, development of time-resolved methodologies, such as time–intensity (Echols *et al*., 2003; Eilers and Dijksterhuis, 2004; Le Révérend *et al*., 2008; Morris *et al*., 2009) and temporal dominance of sensations (Bayarri *et al*., 2007; Le Révérend *et al*., 2008; Lenfant *et al*., 2009), and their statistical analysis (Lenfant *et al*., 2009) has greatly boosted our understanding of the phenomena and transformations occurring in the mouth during eating. Since no *in situ* analytical methods are available to understand the physical basis underlying the changes in taste and texture perception, numerical models have been developed to simulate oral processing and evaluate the phenomena that mostly contribute to those changes (Van Der Glas *et al*., 1987; de Loubens *et al*., 2011; le Révérend and Bakalis, unpublished).

In situ studies of aroma during eating appear to be more promising, as gases can be sampled in the nostrils and analysed through spectroscopic techniques, such as atmospheric pressure chemical ionisation mass spectrometry (APCI-MS) or proton transfer reaction mass spectrometry (PTR-MS) (Malone *et al*., 2003a; Poinot *et al*., 2009). Compared with liquids and solids present in the mouth, sampling of inviscid fluids is considerably simpler. In addition, it can be safely assumed that the acquired sample is very representative of the actual gas sensed by the olfactory receptors, thus allowing the release rates of volatile molecules from foods during their oral processing and the kinetics of decay of those molecules from the gas sensed by the olfactory receptors to be

modelled. These kinetics can also be used to validate *in silico* models of food oral processing (de Loubens *et al*., 2011).

Due to the above-mentioned significant difficulties, often unsurpassable to date, associated with precise monitoring of food oral processing *in vivo*, and in an attempt to avoid the numerical complexity and model simplicity of simulation, *in vitro* simulators have also been developed. These will be discussed in the next section.

7.3 INSTRUMENTAL METHODS AND MOUTH SIMULATORS

The first formal introduction of food texture perception into the scientific community dates back to the 1960s, where perception of food was related to its density, viscosity, surface tension "and other physical properties" (Chen, 2009). Since then, food texture analysis has been paramount to the understanding of the mechanical properties of foods, and their links with sensory attributes. Historically, sensory attributes related to texture have been linked to bulk viscosity (Chen, 2009). As such, the first attempts to simulate phenomena occurring during oral processing engaged rheological measurements of bulk phases. One should note, however, that rheology is often limited to specific sensory attributes and the complete picture of sensory perception during eating extends beyond rheological measurements (van Aken *et al*., 2007; Chen, 2009). This can be partially attributed to the wide range of conditions or phenomena experienced during oral processing, and the fact that processing of different foods is very different in nature.

Texture profile analysis was first developed by General Foods (now Kraft) in the 1960s (Friedman *et al.*, 1963; Chen, 2009). Their so-called Texturometer was later adapted to an Instron Universal Testing Machine and subsequently refined by Bourne (1978), but the original experimental concept was maintained. The test involves a double compression of materials and has become essential to the industry standards over the last few decades. Using this technique, a number of textural elements (hardness, chewiness, cohesiveness, viscosity, etc.) have been correlated with the obtained force–displacement data. Another, now classic, example of this era of texture analysis is the work of Shama and Sherman, 1973 and Shama *et al.*, 1973, who correlated the perceived thickness of a variety of foods to bulk rheology for shear rates ranging from $10\,s^{-1}$ to $1000\,s^{-1}$. It should be noted, however, that for non-Newtonian foods, high viscosities as perceived by sensory panels did not correlate very well to bulk viscosity as measured using this method.

It was in the late 1980s when Kokini (1987) correlated thin-film rheology and friction of foods, as they are manipulated within the oral cavity, to creaminess and thickness. This approach has markedly promoted our understanding of the relationship between aspects of sensory perception and lubrication of foods during eating, thus further encouraging the development of predictive capabilities (de Vicente *et al*., 2006; Bongaerts *et al*., 2007). The relevant experimental area of study is tribology.

The term "tribology" originates from the Greek work for "rub" (τρίβω). The method involves measurements of the friction between two surfaces in relative motion separated by a thin film of lubricating material. The field of tribology originated in the development of lubricants, while a recent interest in the areas of biology has been identified (Taylor, 2012). In a typical tribology experiment, lubrication properties are represented with a Stribeck curve, where the measured friction (or traction) is drawn as a function of entrainment speed (see Fig. 7.4). Typically, a Stribeck curve consists of three distinctive areas, corresponding to three different lubrication regimes. At low sliding speeds, friction is dominated by surface-to-surface contacts, and very little, or no, lubricant exists between the surfaces, resulting in the characteristic high values of the friction coefficient typical of the *boundary* regime. With an increase in entrainment speed, fluid is entrapped between the surfaces separating them and results in a decrease in the friction coefficient. This regime is termed *mixed*. Further increase in speed will

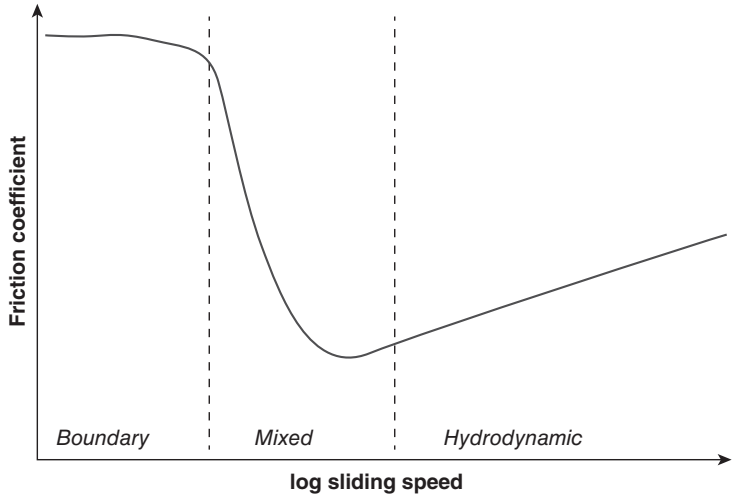


Fig. 7.4 Schematic of a Stribeck curve showing the different lubrication regimes. With kind permission from Springer Science+Business Media: Food Biophysics, Tribology of o/w Emulsions Under Mouth-like Conditions: Determinants of Friction, 2:4, 2007, p. 159, Dresselhuis *et al.*, Figure 1. © Springer Science + Business Media, LLC 2007.

finally move the system into the *elasto-hydrodynamic* regime, where the surfaces become completely separated from each other and the bulk (rheological) properties of the lubricant now determine the measured friction (Czichos, 1978). For time-dependant materials, such as unstabilised emulsions, tribological experiments under constant speeds are suggested instead. These would allow investigation of these systems under "mouth-like" conditions.

The tribological behaviour of emulsions has been investigated by Malone *et al*. (2003b). They reported that systems with oil content below 15% yielded curves that overlapped with the curve of pure water, indicating a water-continuous contact under all stages of lubrication. On the other hand, a 55% oil-containing emulsion exhibited the highest score in terms of fattiness (speeds >100 mm/s). It is important to keep in mind that flavours and volatiles present in the oil phase can affect the overall sensorial perception of the food, even if the lubrication sensation remains the same. In the same work, the authors further demonstrated good correlation (with a factor of 0.99) between sensory perception (oral slipperiness) and friction coefficient in the mixed lubrication regime, suggesting that it is this region where phenomena occurring between the tongue and palate during oral processing/assessment are best mimicked.

When designing tribology-based experiments, careful selection of the relevant surfaces is crucial, as it may play a paramount role in the final results. Typically for food-related experiments, soft surfaces such as poly-dimethylsiloxane (PDMS) or silicone are used to mimic the soft oral surfaces (de Vicente *et al*., 2006; Bongaerts *et al*., 2007). Comparison between "artificial" and "real" materials has been performed by Dresselhuis *et al*. (2008), who conducted a series of identical experiments using either PDMS or biological tissue (pig's tongue). The authors indeed found significant differences between the actual oral surfaces and PDMS, especially in terms of roughness and hardness. However, it is widely understood that the use of biological material as mouth analogues has considerable practical, and potentially ethical, constraints. As such, an artificial surface would be preferential, although in need of some improvement. Furthermore, it has been recently suggested that good correlations can still be obtained between sensory and friction measurements even when relatively rough PDMS surfaces are used (Ranc *et al*., 2006). It comes as no surprise, therefore, that PDMS is still the material of choice in many food-related tribological measurements.

Over the past few years, significant effort has also been devoted to developing *in vitro* mouth simulators for solid products (van Ruth and Roozen, 2000; Salles *et al*., 2007; Woda *et al*., 2010; Mills *et al*., 2011; Benjamin *et al*., 2012). Typically, samples are introduced into

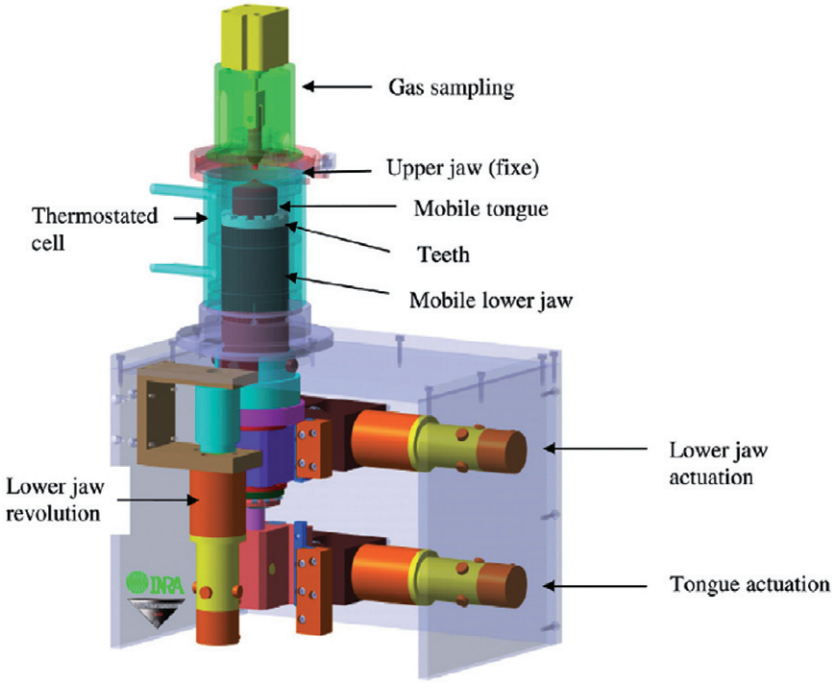


Fig. 7.5 Schematic diagram of a mouth model. Reprinted from Salles *et al.*, (2007) Development of a chewing simulator for food breakdown and the analysis of in vitro flavor compound release in a mouth environment. *Journal of Food Engineering* 82, 189–198. Copyright 2007 with permission from Elsevier.

the simulated oral cavity, along with an amount of simulated saliva, added either in batch or continuous pattern. Mastication is then replicated by the application of mechanical forces, exerted with the aid of a stirrer or a compression mechanism, or a combination of both. The objective of many existing mouth models is to either investigate structural breakdown of foods and bolus formation, or to characterise flavour/aroma release profiles. In the latter case, the model is often connected to a gas chromatograph (GC) or gas chromatograph-mass spectrometer (GC/MS).

An interesting mouth model has been developed by Salles *et al.* (2007), who used biocompatible inert materials and 3D engraved teeth for its construction (see Fig. 7.5). This simulator allows control and setting of some mastication parameters (biting force and frequency, shearing angle, saliva flow rate, etc.) throughout the process simulation, and it appears to successfully represent realistic chewing conditions.

The phenomena occurring during eating are not only complex to mimic, but also convoluted to understand, as there are interactions between the food and the oral surfaces, as well as feedback mechanisms as a response to different foods. Efforts have rightly aimed at developing models to study specific types of food (e.g. liquids, soft solids,

solids) under a specific scope (e.g. fracture, bolus formation or aroma release), because the development of a unified model seems at present an unrealistic target. One of the typical constraints when considering the accuracy of mouth simulators is the limited validation of the models with sensory data. Overall, *in vitro* modeling is not only an active area of research, but also a very challenging one. Considering the need of the industry to obtain predictive capabilities in the field, this is bound to grow over the years.

7.4 INTERACTIONS OF FOODS (EMULSIONS, SOFT SOLIDS, HARD SOLIDS) WITH THE ORAL SURFACES

As food is received in the mouth, it contacts the surfaces and contents of the oral cavity, where it is further processed and appreciated. Many of the oral processes described above, as well as sensory perception of the food, occur as food material interacts with the oral surfaces and the layer of saliva and mucous that overlays them (van Aken *et al*., 2007). Interactions between consumed foods and the oral ingredients are, therefore, determinant factors of the eating process and acquired pleasure, and will be discussed in this section.

7.4.1 Saliva

Saliva is a complex, heterogenous liquid with a multifunctional role in human oral physiology (e.g. speech, lubrication and digestion of foods, maintenance of oral health) (Humphrey and Williamson, 2001; van Aken *et al*., 2007). Saliva consists of roughly 99.5% water, 0.3% protein and 0.2% inorganic and trace substances, including electrolytes, mucus, glycoproteins, antibacterial compounds and enzymes (van Nieuw Amerongen *et al*., 2004). The exact composition and physicochemical characteristics of saliva, including pH and rheological properties, change during mastication and with the condition of the mouth (e.g. health). The natural pH of saliva is in the range of 6–7, with an average of 6.75, indicating a slightly acidic character (Humphrey and Williamson, 2001). In addition, saliva is considered to be a non-Newtonian fluid, showing a characteristic decrease in viscosity as shear rate increases, which has been attributed to the presence of glycoproteins, e.g. mucins (van Aken *et al*., 2007).

Saliva is secreted into the mouth by three pairs of major salivary glands (parotid, sublingual and submandibular glands) and a number of minor ones (Dodds *et al*., 2005). The rate of secretion by each gland differs under different stimulations. Table 7.1 shows typical average contributions of each gland to the total salivary flow rate reported for four different stimulatory conditions: sleep, no stimulation, mechanical

Table 7.1 Average percentage contribution by each gland under different stimulation. Reprinted from Aps and Martens (2005) Review: the physiology of saliva and transfer of drugs into saliva. *Forensic Science International* 150(2-3), 119–131. Copyright 2005 with permission from Elsevier.

Salivary glands	Sleep (%)	No stimulation (%)	Mechanical stimulation (%)	Citric acid stimulation (%)
Parotid glands	0	21	58	45
Submandibular gland	72	70	33	45
Sublingual glands	14	2	2	2
Minor glands	14	7	7	8

stimulation and citric acid stimulation (Aps and Martens, 2005). The gland size also affects secretion rate. Ono *et al*. (2006), measured the size of glands using MRI, and found that rate of saliva secretion from larger glands is higher than that of the smaller-sized glands. Jenkins (1978) reported average adult flow of unstimulated saliva of 0.43 mL/min, with increasing potential at 0.77–4.15 mL/min when stimulation occurs.

Saliva has an important role in oral processing of foods, as it plays a part in lubrication, digestion, buffering, sensory perception, flavour release and formation of food bolus. It is, for example, the lubricating effect of saliva that allows food transformation into swallowable masses and permits smooth transfers in the mouth with minimal irritations to the oral surfaces (Chen, 2009). Saliva also contains the enzyme α-amylase, a catalyst for the hydrolysis of starch. This initiates starch digestion during chewing and results in the consequent decrease in food viscosity. As food passes through the stomach, starch digestion is halted due to inactivation of the enzyme by the strong acidic environment, and it resumes in the small intestine, where most of starch decomposition occurs (Chen, 2009).

Overall, the numerous interactions of food with saliva during oral processing stimulate mixing and formation of different structures, facilitate food transformations and promote changes in the physical and chemical properties of food. The resulting viscosity is likely to affect texture evolution, flavour release profiles and the general perception of the food being processed. Notably, the electrostatic effect of saliva on consumed emulsions may additionally cause droplet flocculation, and this is expected to further impact upon the mechanical and sensory characteristics of the consumed food. Saliva-induced emulsion flocculation has been investigated *in vitro* and *in vivo* (Vingerhoeds *et al*., 2009; Vliet *et al*., 2009). The electrostatic salivary properties have been considered as part of a range of induced mechanisms occurring during oral processing, including depletion flocculation, and electrostatic and van der Waals interactions. The level of the droplets' surface charge and charge distribution have been identified as factors with a strong influence on the mechanism of emulsion flocculation during eating. For

example, in the work of Silletti *et al*. (2007), no flocculation was observed in highly negatively charged O/W emulsions stabilised by sodium dodecyl sulfate (SDS). Conversely, weakly negatively charged emulsions (stabilised by beta-lactoglubulin or Tween 20) flocculated reversibly, indicating a strong element of depletion flocculation in the flocculating mechanism (Silletti *et al*., 2007). While similar observations have also been reported for sodium caseinate emulsions mixed with saliva, as well as with pig gastric mucin, it was also remarked that the concentration of model mucin required to induce flocculation was significantly higher (0.4 wt%) than the average concentration of saliva (0.02 wt%). In their recent work, Silletti *et al*. (2007) proposed that bridging flocculation could also contribute to saliva-induced emulsion flocculation. In particular, they reported formation of irreversible flocculation when positively charged emulsions were mixed with saliva, suggesting bridging of droplets facilitated by binding of the negatively charged mucins onto the adsorbed protein layer on the droplets' surface Silletti *et al*. (2007). In addition, Yao *et al*. (2003) indicated that, additionally to mucins, more salivary components, such as cystatins and serum albumin, are negatively charged and might have a role in the observed flocculation phenomenon.

7.4.2 Mucous layer

Oral surfaces are naturally coated by a mucus layer with a thickness of around 40 μm. The coating's thickness can be determined by application of filter paper strips on the mucus layer and subsequent measurement of the volume of trapped saliva over the relevant oral surface area (Wolff and Kleinberg, 1998). The mucus coating contains 95% water and 0.5–5% mucins (van Aken *et al*., 2007). The roles of these mucins in the human oral cavity are multifold, including tissue coating of oral hard and soft tissues, lubrication of food and oral surfaces, and modulation of oral flora (Tabak, 1990).

From a chemical perspective, mucins are high-molecular-weight glycoproteins with a characteristic peptide core (apomucin) rich in serine, threonine and proline residues, and carbohydrate side chains (oligosaccarides). There are two distinct structures of mucins present in the mucus layer: MG2, the low-molecular-weight species, and MG1, essentially an assemblage of multiple subunits covalently linked to each other, yielding a suprastructure with an aggregate molecular weight in excess of 1 million.

As food reaches the mucosal layer, it is exposed to the pertinent mucins, and the strength of the consequent interactions often determines the residence time of food ingredients, including aroma and flavour compounds, in the mouth. Interestingly, certain polymeric

materials (e.g. chitosan, Carbopols®, or sodium alginate) have the ability to "stick" onto mucosal layers, and therefore offer the attractive perspective of engineering foods with the desired mucoadhesive properties and enhanced aroma and/or flavour perception (Le Révérend *et al*., 2010).

Mucoadhesive biopolymers have gained substantial popularity within the area of pharmaceutical research, due to their potential to: (1) increase the drug's residence time (Andrews *et al*., 2009) and (2) maintain a high concentration gradient of drug across the epithelium (Govender *et al*., 2005) in the mucosa of the digestive tract. In addition, mucoadhesives have been linked with increased permeability of the epithelial wall caused by a series of sequential steps involving dehydration of the mucus layer, "shrinking" of epithelial cells, opening of tight junctions and consequent creation of supplementary paracellular pathways for drug absorption (Lehr, 2000).

There are, overall, five types of chemical interaction occurring between mucoadhesives and mucous layers: (1) *ionic* bonds (two oppositely charged entities attracted to each other by electrostatic forces); (2) *covalent* bonds (atoms sharing electrons in such a way as to fill the incomplete orbitals and form stable chemical compounds); (3) *hydrogen* bonds (electromagnetic attractions, weaker than ionic or covalent bonds, between a hydrogen atom and electronegative atoms such as oxygen, nitrogen or fluorine); (4) *van der Waals* interactions (weak dipole–dipole and dipole-induced dipole attractions in polar molecules and dispersion forces with non-polar substances) and (5) *hydrophobic* bonds (attractions between different non-polar groups present in an aqueous solution resulting in localised exclusion of water) (Smart, 2005).

7.4.2.1 *Mechanisms of adhesion*

Six distinctive mechanisms of mucoadhesion have been described in the literature to date, namely electronic, diffusion, adsorption, mechanical and wetting mechanisms (Peppas and Sahlin, 1996; Ahuja and Khar, 1997; Smart, 2005). Each type has its limitations and, not surprisingly, actual adhesion in the body typically involves a combination of mechanisms.

Electronic adhesion occurs when electrons are transferred across the mucin/polymer interacting interface to form an electrical double layer. On the other hand, diffusive adhesion is a time-dependent process defined by diffusion of mucoadhesive polymer chains into the glycoprotein chain network of the mucous layer (Andrews *et al*., 2009). The resulting binding strength depends on the diffusion coefficient and contact time between the two surfaces and it is determined by the

degree of crosslinking (Dodou *et al*., 2005). The resulting bond is said to have a semipermanent character (Smart, 2005).

Adsorption is possibly the main contributor in human mucoadhesion mechanisms, resulting from the combinative effect of hydrogen bonding and attractive van der Waals forces between the interacting entities. Further, (muco) adhesion triggered by the interlocking of liquid adhesive into irregularities on a rough (mucosal) surface is characteristic of the mechanical mechanism. Rough surfaces provide wider contact area between adhesive material and the interacting mucin surface, resulting in increasing of viscoelastic and dissipation of energy during joint failure (Derjaguin *et al*., 1977).

Wetting adhesion benefits from the ability of certain mucoadhesives to spontaneously spread onto the mucin surface. A material's spreading properties can be evaluated based on the spreading coefficient (S_{AB}, Equation 7.1).

$$S_{AB} = \gamma_B - \gamma_A - \gamma_{AB} \tag{7.1}$$

where γ_A is the surface tension of liquid A, γ_B is the surface tension of solid B and γ_{AB} is the interfacial energy between the solid and liquid. A positive value of S_{AB} represents spontaneous spreading.

7.4.2.2 *Steps of interaction between mucoadhesive and mucous layer*

Typically, mucoadhesion involves two stages: contact and consolidation (Wu, 1982). Contact describes the process where the mucoadhesive materials are positioned in close proximity (wetting) with the mucous layer (see Fig. 7.6). This stage is promoted by the physical motions (in the macroscale) occurring during the eating process, which encourage contact between polymers and the mucous layer (Smart, 2005). When in contact, polymers will allow some interaction with the mucins and deposition of particles that have adhesive properties onto the targeted area (mucus layer). Consolidation succeeds contact and involves various physicochemical interactions between the interacting areas necessary to acquire a strong or prolonged adhesion. The stages of adhesion are depicted in Fig. 7.6, while Fig. 7.7 describes some scenarios where mucoadhesion can occur.

The main determinant of the level of interpenetration and entanglement of binding during mucoadhesion, and hence the strength of bonding, is the polymer's molecular flexibility (related to the molecular weight) (Smart, 2005). Other factors that impact on the adhesive process and its effectiveness, include environmental conditions (pH, ionic strength of surrounding media), especially in the case of polymers containing ionisable groups (Gu *et al*., 1988), as well as contact time and spatial scale of contact.

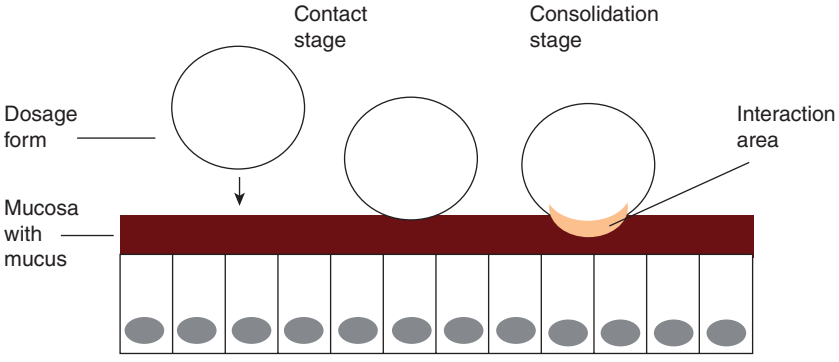
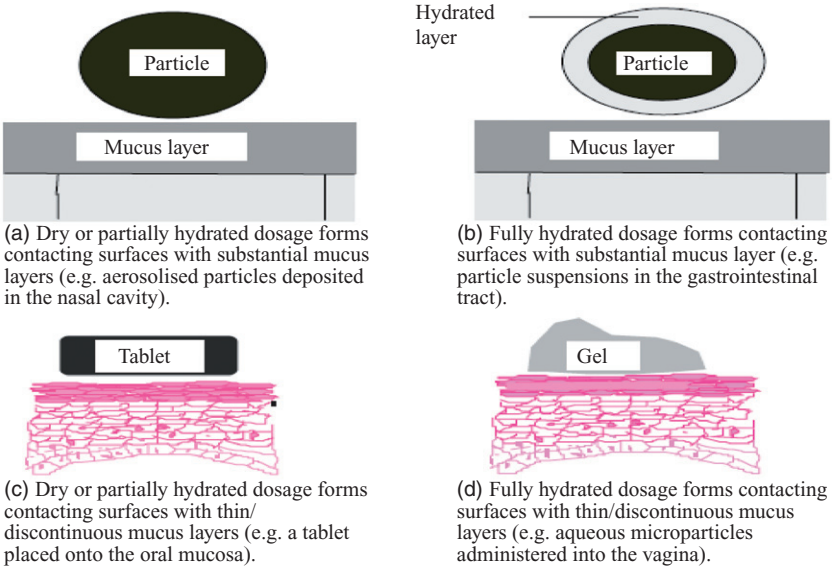


Fig. 7.6 Contact and consolidation stages of mucoadhesion. Reprinted from Smart (2005) The basics and underlying mechanisms of mucoadhesion. *Advanced Drug Delivery Reviews* 57, 1556–1568. Copyright 2005 with permission from Elsevier.



(a) Dry or partially hydrated dosage forms contacting surfaces with substantial mucus layers (e.g. aerosolised particles deposited in the nasal cavity).

(b) Fully hydrated dosage forms contacting surfaces with substantial mucus layer (e.g. particle suspensions in the gastrointestinal tract).

(c) Dry or partially hydrated dosage forms contacting surfaces with thin/discontinuous mucus layers (e.g. a tablet placed onto the oral mucosa).

(d) Fully hydrated dosage forms contacting surfaces with thin/discontinuous mucus layers (e.g. aqueous microparticles administered into the vagina).

Fig. 7.7 Some scenarios where mucoadhesion may occur. Reprinted from Smart (2005) The basics and underlying mechanisms of mucoadhesion. *Advanced Drug Delivery Reviews* 57, 1556–1568. Copyright 2005 with permission from Elsevier.

Detachment of adhered material would normally initiate at the weakest bound position of the interacting mucin/poymer surface, and therefore, durability of the adhesive joint will effectively depend on the cohesive nature of the weakest region (Smart, 2005). When the mucoadhesive material is overhydrated, it will be easily removed by forming slippery mucilage in between the mucoadhesive material and mucin glycoprotein (Chen and Cyr, 1970). Therefore, controlling the rate and

extent of hydration between the mucoadhesive material and mucus layer offers a means of prolonging adhesion, as required. Indeed, some strategies such as crosslinking (Hagerstrom and Edsman, 2001) and introduction of hydrophobic groups (Inoue *et al*., 1997) have already been tried towards this goal.

7.5 HOW COMBINING FOOD ORAL PROCESSING AND FOOD MICROSTRUCTURE HELPS MANIPULATE SENSORY PERCEPTION: THE CASE OF CHOCOLATE

Chocolate is an interesting food when it comes to oral processing, due to its microstructure of crystalline fat, embedded sugar and cocoa powder (Le Révérend *et al*., 2008). The melting point of the continuous cocoa butter (32 °C) is just below that of the mouth's temperature (35 °C) (Wille and Lutton, 1966), which confers to chocolate the melting profile that is necessary for its well-known sensory appeal (Ollivon, 2004).

It has been recently highlighted that individual consumers may have different oral processing behaviours during chocolate consumption (Carvalho-da-Silva *et al*., 2011). Using electromyography and electroglottography to monitor chocolate consumption at a physiological level, the authors were able to identify three characteristic "chocolate eater" profiles: "thorough chewers", "fast chewers" and "suckers". It is also interesting to note that this behaviour is not influenced by chocolate textural properties, and instead it seems to be intrinsic to the person.

In order to manipulate the release of flavour compounds from the chocolate matrix to the taste and flavour receptors, controlling melting rate is the most obvious feature, as transport is facilitated when the matrix viscosity drops (Gady *et al*., 2008). In the case of "suckers" (that let chocolate melt in their mouth) a way to control the melting rate is to maximise the area in contact with the oral cavity. This was recently demonstrated by Lenfant *et al*. (2013), who showed that chocolate pieces with the highest surface area in the contact with the mucosa (*AC*) compared to their volume (*V*) was a good predictor of the oral performance of a chocolate piece when compared with sensory data. The *AC/V* ratio was found to correlate significantly well with the perceived melting attribute, as well as with the intensity of cocoa flavour perceived. Such transformations in the mouth must arise from the quality of heat transfer between the mouth (heat source) and the chocolate (heat sink). This ought to be linked with phase change models that have been developed for chocolate processing purposes (Tewkesbury *et al*., 2000; Padar *et al*., 2008; Le Révérend *et al*., 2009).

Another way to control the sensory characteristics of chocolate is to control not only its macrostructure but also its microstructure. Haedelt *et al*. (2007) showed that by changing the gas used to sparge chocolate to include air bubbles, one could modify its textural and flavour properties. Nitrous oxide and carbon dioxide were found to be more soluble in the matrix than argon and nitrogen. This affects the structure of the chocolate, leading to larger bubbles being formed when using more soluble gases, thus making the resulting specimens less hard and creamy than their denser counterparts, made with less soluble gases. Also, nitrous oxide led to higher aroma perception.

Finally, in the context of growing worldwide obesity, the manufacturing of low-calorie chocolate seems an interesting and relevant challenge. This has been partially tackled by using artificial sweeteners, but in order to reduce both fat and sugar at the same time the use of inert filler particles seems the most attractive route. Recently, Norton *et al*. (2009) presented the use of margarine-like technology to control the interfaces of a water-in-cocoa-butter emulsion that could be used to partially reduce fat and thus calorie content. When taking into account that it is the tastant concentration in the continuous phase that controls the taste properties of hydrocolloids (Malone *et al*., 2003a; Goh *et al*., 2010), this reduction by using inert fillers could also be applied to sugar.

7.6 CONCLUSIONS

Research in several discipline areas, e.g. molecular biology and nutritional medicine, is now providing a deeper scientific understanding of the role and contribution of food consumption and nutrition in both health and disease. Mirroring these improvements in scientific understanding of food and health, there is an equally significant increase in consumer awareness: in many cases this leads to eating a more healthy diet.

However, for various reasons, many consumers find it difficult to adopt a healthy diet and lifestyle and are attracted to the pleasure, convenience and indulgence given by certain foods, despite being aware of the negative contributions to their health and wellbeing. In the EU, food-related diseases have reached epidemic levels, with obesity alone being responsible for 10–13% of the deaths, according to the World Health Organisation. Thus, there is a growing need to intelligently engineer food structures that deliver a healthy and nutritionally balanced diet, but still meet all of the consumer requirements and demands of quality, convenience and eating enjoyment/pleasure. There are clear opportunities enabling collaboration and integration

across disciplines that will develop food structures to deliver both the nutritional and the organoleptic qualities that the modern consumer demands.

In the context of (bio)chemical applications and processes, scientists and technologists have been highly successful in developing the scientific and engineering principles required for manufacturing bulk chemicals: *products that are specified largely by chemical composition*. However, these chemical engineering principles have not been fully exploited in the design of structured foods (typically multicomponent and multiphase), where the end-user (the consumer) benefit of the product is specified by a wide range of functions, such as texture, convenience, ease of preparation, and physical, chemical and microbiological stability, as well as nutritional properties. In order to achieve this, one would require a fundamental understanding of the interactions between food formulation, processing and phenomena occurring during eating. This knowledge would then enable the food engineer to step backwards and design the required structures and chemical compositions to achieve the desired food product.

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