

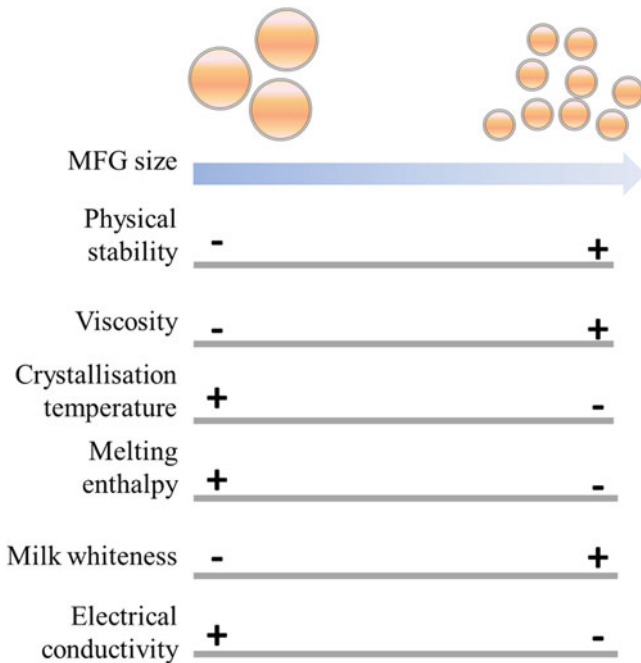
## Chapter 6

# Effect of Milk Fat Globule Size on Physical Properties of Milk

Within the wide size range of MFG, the smallest globules are approximately 100-fold smaller in diameter compared to the largest ones. For the same bulk volume of fat, milk, with smaller MFG will have a higher total number of MFG. Within these milks, the smaller MFG will tend to have greater surface curvature, and a larger surface area/volume ratio, compared to larger MFG. These differences can give rise to marked differences in the physical properties of MFG size-differentiated milk and milk fat as summarised in Fig. 6.1.

### 6.1 Physical Stability

Bovine milk, in which fat globules are dispersed in the continuous phase of milk plasma containing casein micelles, serum proteins, sugars and minerals, can be considered as both a colloidal suspension and an oil-in-water emulsion. Within the emulsion, the MFGM maintains the integrity of the lipid droplets and helps to protect them from destabilisation (Walstra et al. 1999). However, as a natural oil-in-water emulsion, milk is thermodynamically unstable and readily subject to various forms of physical instability over time, leading to changes of structural organisation or spatial distribution of MFG. These instability mechanisms include gravitational separation, droplet aggregation, flocculation, coalescence, and partial coalescence, which are governed by three colloidal interactions, i.e. van der Waals attractions, electrostatic repulsion and steric stabilisation (Huppertz and Kelly 2006). Creaming phenomena can be prevented by reducing the emulsion droplet size. Indeed, for nanoemulsions (i.e. below 200 nm) in general, Brownian motion can be sufficient to overcome the influence of gravitational force (Tadros et al. 2004; Mason et al. 2006; McClements and Rao 2011), thereby providing enhanced long-term physical stability. McClements (2011) calculated that creaming becomes negligible if emulsion droplet size is below 10 nm. However, particles in nanoemulsions can be subjected to sedimentation if



**Fig. 6.1** Illustration of impact of milk fat globule size on selected fundamental properties (“+”: increased; “-”: decreased)

they are coated by a thick adsorbed protein layer to such an extent that their overall density is higher than that of water (McClements 2011). Destabilisation by flocculation and coalescence can also be reduced in nanoemulsions, due to enhanced steric stabilisation (Anton et al. 2008; McClements and Rao 2011; Tadros et al. 2004). Whilst these are well-founded principles with respect to model emulsions, there is little information about the physical stability of native MFG at the nanometric-size scale. However, it is well established that homogenised milks (down to about 0.4  $\mu\text{m}$ ) are relatively stable against creaming and it is reasonable to project that the physical stability of MFG may also be enhanced in nanoemulsions.

Milk fat globule size greatly impacts on the formation of milk fat clusters, as in creaming and cold agglutination, which in turn, affects the physical stability of milk and dairy products. In general, the smaller the fat globule size, the more stable it is. Hence, drinking milks are commonly homogenised to reduce the size of milk fat globules (below 1  $\mu\text{m}$ ) to achieve the greater stability and shelf-life. Large MFG size accelerates the creaming. Furthermore, large MFG tend to have protruding fat crystals, which facilitate partial coalescence (Walstra 1995). Large MFG size, hence, is undesirable in drinking milks but are preferred in butter manufacture, where small MFG in homogenised cream may lead to inefficiencies in the churning process (Walstra et al. 2005).

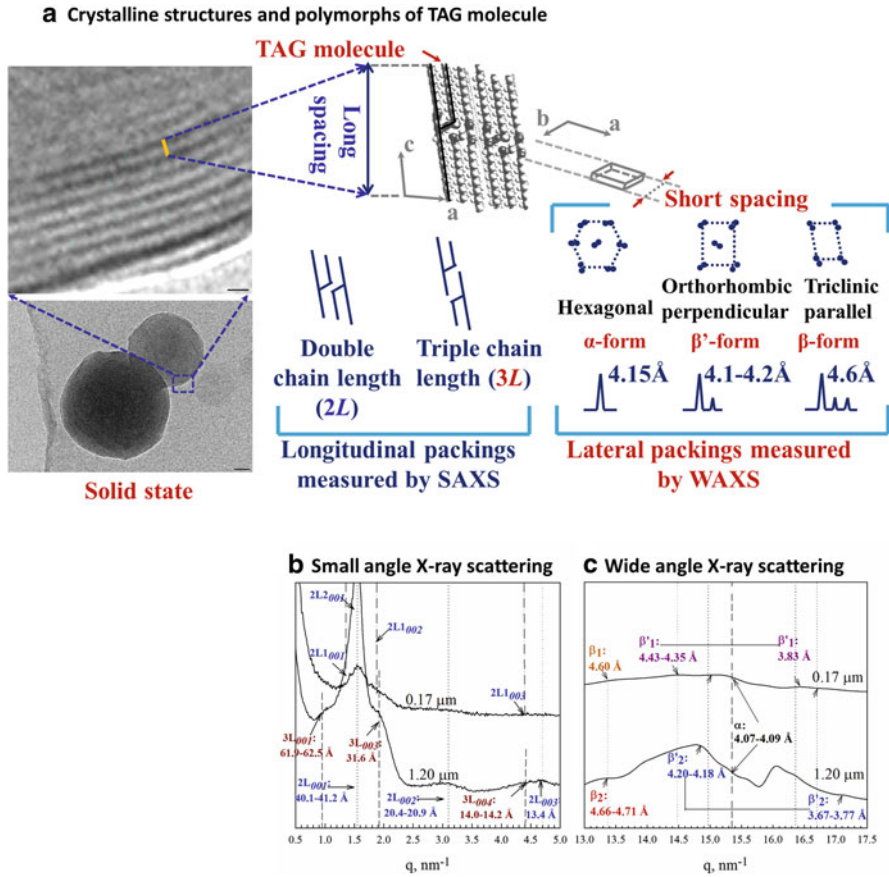
Milk fat globule size is also considered to influence the creaming rate in cold raw milk, through its influence on cold agglutination. This occurs in milk due to the presence of agglutinin, a cryoglobulin-lipoprotein complex (Walstra 1995). On cooling raw milk, cryoglobulins are precipitated and coat the MFG, causing them to aggregate and form large floccules, which then rise as a creaming layer. Since surface area is greater with smaller fat globules, more agglutinin is required to envelop them completely, rendering them inherently more stable to cold agglutination (Walstra et al. 2005).

## 6.2 Viscosity

Viscosity of milk and dairy emulsions is also dependent on MFG size. In homogenised milk, the higher the homogenising pressure, the higher the viscosity. When milk was homogenised from 70 to 245 bar, there was a corresponding increase in viscosity from 7.1 to 15.0 % (Kessler 1981). It was also reported that smaller MFG size causes a slight increase in (apparent) viscosity (Long et al. 2012; Truong et al. 2014a; Kietczewska et al. 2003). A decrease in MFG size of 3.3 % fat milk from 2.7 to 1.0  $\mu\text{m}$  resulted in corresponding higher viscosities (1.8–1.96 mPa s) (Kietczewska et al. 2003). Similar tendencies were noted in dairy-based emulsions (10–36 % milk fat) with much lower size range (0.2–1.3  $\mu\text{m}$ ). When emulsion size decreased from 1.2 to 0.2  $\mu\text{m}$ , the apparent viscosities of the dairy-based emulsions increased in the range of 8–15 mPa s at a shear rate of 5.6  $\text{s}^{-1}$  (Truong et al. 2014a). For a high fat containing emulsions (36 % milk fat) Long et al. (2012) reported a higher apparent viscosity (0.852 Pa s) for the smaller size emulsion (0.415  $\mu\text{m}$ ) compared to the bigger size (1.291  $\mu\text{m}$ ; 0.398 Pa s). The increase in emulsion viscosity of milk and dairy emulsions with decreasing MFG size can be partly explained by stronger colloidal repulsion and monodispersed close packing caused by narrow size distribution and smaller particle size (Pal 1996).

## 6.3 Crystallisation and structural properties

The main components of MFG are TAG, which exist in hundreds of molecular species. The configuration of TAG molecules in their solid state can be described in two dimensions as presented in Fig. 6.2a. The longitudinal stacking (long spacing) is the alignment of repetitive patterns of TAG side-by-side, either in two chain length (2L) or higher (triple 3L, quartet 4L etc. chain length configuration). The cross-sectional view, regarded as the short spacing, is associated with the structural arrangement of the TAG side chains. They are  $\alpha$ ,  $\beta'$ , and  $\beta$  polymorphs having hexagonal, orthorhombic and perpendicular, and triclinic parallel chain packing, respectively (Chapman 1962).



**Fig. 6.2** Schematic illustration of TAG crystalline conformation (a) and impact of droplet size on crystalline packing longitudinally (b) and laterally (c) in olein emulsions. The 3L structure was vanished in the nano-sized emulsion (0.17  $\mu\text{m}$ ) versus the control (1.20  $\mu\text{m}$ ). The emulsion was prepared from 10 % w/w olein fraction (fractionated from AMF at 21  $^{\circ}\text{C}$ ) emulsified with 1 % w/w sodium caseinate (b and c: Reprinted from Truong et al. (2015), Copyright 2015, with permission from Elsevier)

Each TAG has its own melting point and inter-solubility with other TAGs, which results in very complex overall crystallisation and structural properties of milk fat. Upon crystallisation, individual TAG of milk fat can form different polymorphs, depending on previous thermal conditions applied such as cooling/heating temperature and rate. Previous studies on the crystallisation of bovine MFG in cream (natural and recombined) and milk reported the complexity in crystallisation and structural behaviour of milk fat crystals in the dispersion state (Table 6.1). Typically, milk fat crystals in globules are of mixed types of  $\alpha$ ,  $\beta'$  and  $\beta$  depending on rate of cooling (Lopez et al. 2002, 2007).

**Table 6.1** Influence of fat globule size on crystalline structures and polymorphs of milk lipids

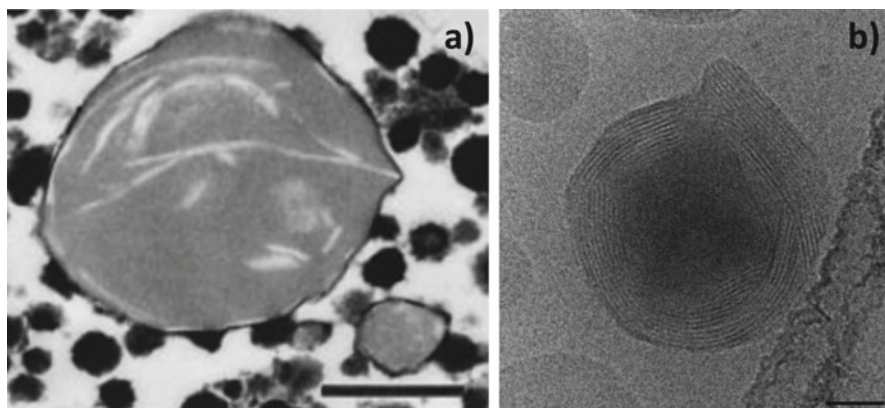
Composition	Droplet size(s)	Cooling condition <sup>a</sup>	T <sub>c</sub> <sup>b</sup>	Longitudinal packing	Lateral packing	References
Native fat globules (small size fraction)	$D_{43}$ 0.93 $\mu\text{m}$	0.5 °C min <sup>-1</sup> (60 to -8 °C)	20 °C	2L (40.6 Å)		Michalski et al. (2004)
			-8 °C	3L <sub>1</sub> (70.7 Å), 3L <sub>2</sub> (64.2 Å)+2L (39.3 Å)		
Native fat globules (large size fraction)	$D_{43}$ 1.75 $\mu\text{m}$	1.0 °C min <sup>-1</sup> (60 to -8 °C)	15 °C	3L (70.6 Å)	$\alpha$	
			-8 °C	2L+3L	$\alpha$ +sub $\alpha$	
			20 °C	2L (41.1 Å)		
Fresh cream	$D_{43}$ 7.15 $\mu\text{m}$	0.5 °C min <sup>-1</sup>	-8 °C	2L (40.8 Å), 3L <sub>1</sub> (68.5 Å), 3L <sub>2</sub> (61.2 Å)		
			25 °C min <sup>-1</sup>	5 °C	3L+2L+3L	$\alpha$ + $\beta'$
Milk fat emulsion (AMF+ $\beta$ -lg)	0.38, 0.45, 0.67, 1.25 $\mu\text{m}$	1 °C min <sup>-1</sup>	-7 °C	3L (72 Å)	$\alpha$	Lopez et al. (2002)
Milk fat emulsions	$d_{0.5}$ 0.17, 0.55, 1.45 $\mu\text{m}$	Steady state	4 °C	3L (56.6 Å), 2L (39.5 Å)	$\beta_1$ + $\beta_2$ + $\beta'_1$ + $\beta'_2$	Bugeat et al. (2011)
			4 °C	2L (41.8 Å)	$\beta_1$ , $\beta'_1$ , $\beta'_2$	
UFA enriched emulsion	Mode 1.2 $\mu\text{m}$	Steady state after cooling at 1.0 °C min <sup>-1</sup>	4 °C	2L (41.1 Å)	$\beta_1$ , $\beta'_1$ , $\beta'_2$	Truong et al. (2015)
				2L (41.5 Å)	$\beta_1$ , $\beta'_1$ , $\beta'_2$	
				2L (40.8 Å)	$\beta_1$ , $\beta'_1$ , $\beta'_2$	
				2L (41.1 Å)	$\beta_1$ , $\beta'_1$ , $\beta'_2$	
Olein emulsion <sup>c</sup>	1.2 $\mu\text{m}$	Steady state after cooling at 1.0 °C min <sup>-1</sup>	4 °C	3L (61.9 Å)+2L (40.5 Å)	$\alpha$ , $\beta'_1$ , $\beta'_2$ , $\beta_2$	
				2L (40.1, 44.3 Å)	$\alpha$ , $\beta'_1$ , $\beta'_2$ , $\beta_1$	
				3L (62.5 Å)+2L (40.5 Å)	$\alpha$ , $\beta'_1$ , $\beta'_2$ , $\beta_2$	
				2L (40.5, 45.3 Å)	$\alpha$ , $\beta'_1$ , $\beta'_2$ , $\beta_1$	

<sup>a</sup>Cooling condition includes cooling rate and temperature range used<sup>b</sup>Crystallisation temperature where the crystalline packings were detected<sup>c</sup>Emulsions prepared from fractionated milk fats (stearin-enriched and olein-enriched)

Fat globule size is also one of the factors influencing the crystallisation and structural characteristics of native MFG (Lopez et al. 2007; Michalski et al. 2004) as summarised in Table 6.1. Using microfiltration to obtain small (1–3  $\mu\text{m}$ ) and large (5–7  $\mu\text{m}$ ) globule size fractions, Michalski et al. (2004) reported that crystallisation was delayed with smaller MFG ( $D_{43}$  0.93  $\mu\text{m}$ ) compared to large MFG ( $D_{43}$  7.15  $\mu\text{m}$ ). The latter forms 3L structure at higher temperature (13 °C) than the former (9 °C). Also, the large size fraction exhibits more 2L structure (2L: 40.8 Å) than its smaller counterpart (2L: 39.3 Å). However, the authors pointed out that no significant discrepancy was found across the size range of native milk fat globules investigated (0.93–7.14  $\mu\text{m}$ ) once the associated cooling rate and thermal history were omitted.

Due to the technical difficulty in separating native MFG into discrete size fractions, few attempts have been made to study the influence of droplet size on crystallisation and structural behaviour of bovine milk TAGs in milk fat emulsion systems. In these systems, anhydrous or fractionated bovine milk fats were used as an oil phase whereas the aqueous phase typically consisted of whey proteins and/or caseins acting as emulsifiers. Generating discrete emulsion droplet size ranges is generally more controllable by varying homogenising pressure and cycles applied to the coarse emulsions during the emulsification process. Furthermore, this method effectively eliminates the compositional variations between small and large MFG in their native form. Using this approach, it was shown that a decrease in droplet size induced a lower crystallisation temperature (Lopez et al. 2002; Truong et al. 2014b), lower solid fat content (Truong et al. 2014b) and smaller melting enthalpy (Bugeat et al. 2011) (Table 6.1). These results were in agreement with studies performed on other fats/oils such as *n*-hexadecane, tripalmitin, tristearin, and trilauroylglycerol (Higami et al. 2003; Bunjes et al. 2000; Dickinson et al. 1991). The tendency of crystallisation temperature to decrease with smaller droplet size can be explained by the increased ratio of droplets to impurities, which are responsible for catalysing or “seeding” crystallisation of individual droplets. An increase in this ratio tends to limit the rate of crystallisation and results in an increasing propensity for supercooling. In nanoemulsions, it is likely that the crystallisation process is further retarded due to the physical constraints of the droplet wall. For example, Bugeat et al. (2011) and Truong et al. (2015) reported the absence of TAG 3L structure in bovine milk enriched unsaturated fatty acid (olein fraction) nanoemulsion (approximately 200 nm) compared to micron-sized emulsions of the same composition (Fig. 6.2b and c), suggesting that confinement of emulsion droplet size constrains and retards the crystallisation and fat crystal growth.

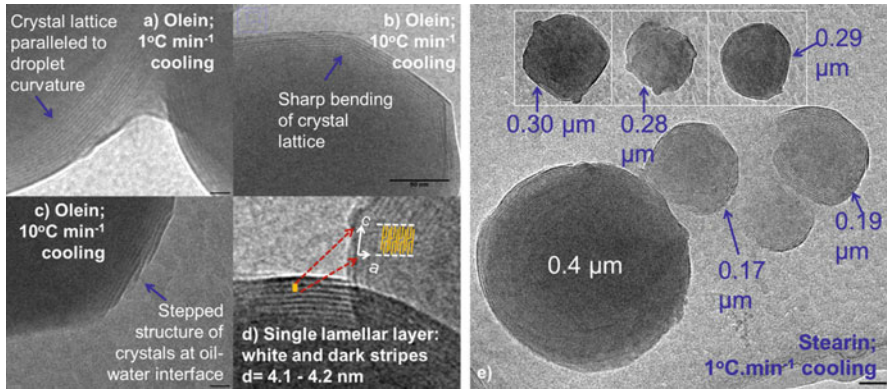
Regarding morphology, milk fat that is crystallised within size-differentiated MFGs exhibits different crystal arrangements and shape, as visualised under polarised light microscopy, depending on MFG size (Lopez et al. 2002). Depending on the microscopic technique used, four main types of crystals in MFG have been proposed based on (1) birefringence of the crystals under polarized light microscopy (Walstra 1967) and (2) location of crystal shell within the fat globule as observed with freeze-fracturing and electron microscopy (Precht 1988). The four main types of crystals observed in cream are: O (tiny crystals interiorly located,



**Fig. 6.3** (a) Presence of needle-like fat crystals inside a bovine MFG captured by Transmission Electron Microscopy (Reprinted from Goff (1997), Copyright (1997), with permission from Elsevier), bar scale: 0.5  $\mu\text{m}$ ; and (b) Arrangement of TAG lamellar layers in both interior and free surface of the stearin-enriched nanoparticle (230 nm) after cooling at very slow rate  $0.1\text{ }^{\circ}\text{C min}^{-1}$  as visualised under cryogenic Transmission Electron Microscopy (Reprinted from Truong et al. (2015), Copyright (2015), with permission from Elsevier), bar scale 50 nm

showing no birefringence), N (“needle-type”: birefringent areas of needle crystals), L (“Layer-type”: needle crystals tangentially located at the outer layer) and M (“mixed type”: combination of L and N types) (Walstra 1967). As illustrated in Fig. 6.3a, needle-fat crystals were detected in interior part of native MFG (Goff 1997). Based on these categories, Lopez et al. (2002) reported that during rapid cooling from  $60$  to  $-8\text{ }^{\circ}\text{C}$ , it is likely that the fat crystals in the smallest fat globules were very small and attributed to type O (very weak birefringence). The largest fat globules held needle-shaped crystals (type N). When native MFG were cooled at slower rate ( $0.5\text{ }^{\circ}\text{C min}^{-1}$ ) the largest globules had a combination of type N and M (layered + needle-shaped crystals) whilst spherulite-shaped crystals were present in the smallest globules (Lopez et al. 2002). A mono-molecular layer about 5 nm in thickness was found to surround the MFG boundary in concentric layers upon crystallization (Precht 1988). For a dairy-based emulsion system, the higher resolution of transmission electronic microscopy revealed that upon very slow crystallisation of TAG ( $0.1\text{ }^{\circ}\text{C min}^{-1}$ ), TAG layers appeared to be arranged into a straight orientation (Fig. 6.3b). The influence of droplet size on crystal morphologies was also demonstrated by Truong et al. (2015) (Fig. 6.4). Here it was apparent that the bent crystals aligned tangentially to the curved interface of micron-sized emulsion droplets were absent from the nanoemulsion droplets. It was suggested that these more typical crystals were unable to form, due to the physical confinement and extreme curvature of the nano-sized droplets (200 nm) (Fig. 6.4). Instead, the crystal lattice within the nano-sized droplets tended to arrange into a straight orientation, causing a deformation of droplets or protruding fat droplet surface (Figs. 6.3b and 6.4).





**Fig. 6.4** Cryogenic Transmission Electron Microscopy micrographs present different arrangements of TAG lamellar layers externally in olein (**a–c**) and stearin (**d**) nanoemulsion (200 nm) at 4 °C after being cooled at different cooling rates (**b** and **c**; Reprinted from Truong et al. (2015), Copyright (2015), with permission from Elsevier). Single lamellar layer (*white and dark thick lines*) about 4.1–4.2 nm, corresponding to the length of TAG longitudinal packing (red arrows), stacked along the periphery of particle (**d**). Polydispersity and various morphologies at different particle sizes in the stearin nanoemulsion (**e**)

## 6.4 Optical Properties

Turbidity of milk is governed by light scattering from milk components, notably fat globules and casein micelles (Goulden 1958; Walstra et al. 2005). Light scattering is stronger with MFG than casein micelles in milk due to a high polydispersity of fat globule size (Walstra et al. 2005). Both fat globule size and fat concentration contribute to light scattering (Goulden 1958; Walstra et al. 2005). Hence, optical properties have been utilised to indirectly estimate MFG size in homogenised milks using spectroturbidimetry (Ashworth 1951; Goulden 1958) and more recently, light scattering (McCrae and Lepoetre 1996; Michalski et al. 2001). On investigating measurement of the optical properties of milk as a means of obtaining reliable MFG size distribution estimates using laser light scattering, Michalski et al. (2002) observed that there was no significant difference in size distribution of natural fat globules using both the corrected (1.458 at 633 nm- and 1.460 at 466 nm-wavelength) and true refractive indices (1.470 and 1.460 at 466 and 633 nm wavelength, respectively). However, using the corrected refractive indices of milk fat improves the peak selectivity in the submicron size range of homogenised milk. These findings appear to indicate at least a small degree of influence of particle size on optical properties of MFG.

The colour and opacity of milk is attributable to both light scattering and absorbance of visible light (Walstra et al. 2005). It is known that homogenised milk appears whiter than raw milk. Sonicated milk, having mean diameter of globule size below



1  $\mu\text{m}$ , is reported to have a significantly higher level of luminosity ( $L^*$ :  $92.37 \pm 0.20$ ) compared to raw milk ( $87.82 \pm 0.18$ ). This was apparently due to an increase in scattering of visible light with smaller fat globules (Fox and McSweeney 1998).

## 6.5 Electrical Conductivity

Milk generally exhibits good electrical conductivity, largely due to the presence of dissociated soluble salts. The presence of milk fat reduces electrical conductivity, due to the poor conductivity of the fat itself, as well as the immobilisation of conducting ions by MFG. Increasing fat content generally leads to a decrease in electrical conductance. It has been observed that at the same level of fat, commercial full fat milk which has a smaller fat globule size, had a higher conductance ( $5.05 \pm 0.03$  mS), than that of raw milk ( $4.85 \pm 0.03$  mS) (Mabrook and Petty 2003). In a separate study, when milk was homogenised, conductance properties such as impedance and admittance of homogenised milk remained unchanged in larger particle size emulsions (1.5–5  $\mu\text{m}$ ; Banach et al. 2008). However, a marked decrease in homogenised milk impedance was observed in milks with a smaller fat globule size (1.07  $\mu\text{m}$ , homogenised at 20 MPa). This observed difference was attributed to the greater disruption and disintegration of casein micelles under the higher homogenisation pressures. It was assumed that during the mechanical size reduction process, part of the colloidal calcium phosphate was dissociated from the micelles and solubilised, leading to an increase in calcium and phosphate contents in milk serum. This contributed to the imbalance of mineral salts in milk, altering its electrical conductivity (Banach et al. 2008). Given that the fat globule membrane in homogenised milks is not of a highly conductive nature, it appears that any differences seen in conductivity between homogenised milks of different emulsion particle size are more likely to be due to the effects of homogenisation on micelle disruption than any direct effect of emulsion droplet size.

## References

- Anton N, Benoit JP, Saulnier P. Design and production of nanoparticles formulated from nano-emulsion templates - a review. *J Control Release*. 2008;128(3):185–99. doi:[10.1016/j.jconrel.2008.02.007](https://doi.org/10.1016/j.jconrel.2008.02.007).
- Ashworth US. Turbidity as a means for determining the efficiency of homogenization. *J Dairy Sci*. 1951;34(4):317–20.
- Banach JK, Żywica R, Kiełczewska K. Effect of homogenization on milk conductance properties. *Pol J Food and Nutr Sci*. 2008;58(1):107–11.
- Bugeat S, Briard-Bion V, Perez J, Pradel P, Martin B, Lesieur S, Bourgaux C, Ollivon M, Lopez C. Enrichment in unsaturated fatty acids and emulsion droplet size affect the crystallization behaviour of milk triacylglycerols upon storage at 4 degrees C. *Food Res Int*. 2011;44(5):1314–30. doi:[10.1016/j.foodres.2011.01.003](https://doi.org/10.1016/j.foodres.2011.01.003).

- Bunjes H, Koch MHJ, Westesen K. Effect of particle size on colloidal solid triglycerides. *Langmuir*. 2000;16(12):5234–41.
- Chapman D. The polymorphism of glycerides. *Chem Rev*. 1962;62:433–56.
- Dickinson E, McClements DJ, Povey MJW. Ultrasonic investigation of the particle-size dependence of crystallization in N-hexadecane-in-water emulsions. *J Colloid Interface Sci*. 1991;142(1):103–10.
- Fox PF, McSweeney PL. *Dairy chemistry and biochemistry*. London: Blackie Academic & Professional; 1998.
- Fredrick E, Van de Walle D, Walstra P, Zijtveld JH, Fischer S, Van der Meeren P, Dewettinck K. Isothermal crystallization behaviour of milk fat in bulk and emulsified state. *Int Dairy J*. 2011;21(9):685–95. doi:[10.1016/j.idairyj.2010.11.007](https://doi.org/10.1016/j.idairyj.2010.11.007).
- Goff HD. Instability and partial coalescence in whippable dairy emulsions. *J Dairy Sci*. 1997;80(10):2620–30.
- Goulden JDS. Some factors affecting turbidimetric methods for the determination of fat in milk. *J Dairy Res*. 1958;25(2):228–35.
- Higami M, Ueno S, Segawa T, Iwanami K, Sato K. Simultaneous synchrotron radiation X-ray diffraction - DSC analysis of melting and crystallization behavior of trioleoylglycerol in nanoparticles of oil-in-water emulsion. *J Am Oil Chem Soc*. 2003;80(8):731–9.
- Huppertz T, Kelly AL. Physical chemistry of milk fat globules. In: Fox PF, McSweeney PLH, editors. *Advanced dairy chemistry volume 2: lipids*, vol. 2. 3rd ed. New York: Springer; 2006.
- Kessler HG. Emulsifying \_ Homogenizing. In: *Food engineering and dairy technology*. Germany: Verlag A. Kessler; 1981. p. 119–38.
- Kietczewska K, Kruk A, Czerniewicz M, Warminska M, Haponiuk E. The effect of high-pressure homogenization on changes in milk colloidal and emulsifying systems. *Pol J Food Nutr Sci*. 2003;12/53(1):43–6.
- Long Z, Zhao MM, Zhao QZ, Yang B, Liu LY. Effect of homogenisation and storage time on surface and rheology properties of whipping cream. *Food Chem*. 2012;131(3):748–53.
- Lopez C, Bourgaux C, Lesieur P, Bernadou S, Keller G, Ollivon M. Thermal and structural behavior of milk fat - 3. Influence of cooling rate and droplet size on cream crystallization. *J Colloid Interface Sci*. 2002;254(1):64–78. doi:[10.1006/jcis.2002.8548](https://doi.org/10.1006/jcis.2002.8548).
- Lopez C, Bourgaux C, Lesieur P, Ollivon M. Coupling of time-resolved synchrotron X-ray diffraction and DSC to elucidate the crystallisation properties and polymorphism of triglycerides in milk fat globules. *Lait*. 2007;87(4–5):459–80. doi:[10.1051/Lait:2007018](https://doi.org/10.1051/Lait:2007018).
- Mabrook MF, Petty MC. Effect of composition on the electrical conductance of milk. *J Food Eng*. 2003;60(3):321–5. doi:[10.1016/S0260-8774\(03\)00054-2](https://doi.org/10.1016/S0260-8774(03)00054-2).
- Mason TG, Wilking JN, Meleson K, Chang CB, Graves SM. Nanoemulsions: formation, structure, and physical properties. *J Phys Condens Matter*. 2006;18(41):R635–66. doi:[10.1088/0953-8984/18/41/R01](https://doi.org/10.1088/0953-8984/18/41/R01).
- McClements DJ. Edible nanoemulsions: fabrication, properties, and functional performance. *Soft Matter*. 2011;7(6):2297–316. doi:[10.1039/c0sm00549e](https://doi.org/10.1039/c0sm00549e).
- McClements DJ, Rao J. Food-grade nanoemulsions: formulation, fabrication, properties, performance, biological fate, and potential toxicity. *Crit Rev Food Sci Nutr*. 2011;51(4):285–330. doi:[10.1080/10408398.2011.559558](https://doi.org/10.1080/10408398.2011.559558).
- McCrae CH, Lepoetere A. Characterization of dairy emulsions by forward lobe laser light scattering - application to milk and cream. *Int Dairy J*. 1996;6(3):247–56. doi:[10.1016/0958-6946\(95\)00008-9](https://doi.org/10.1016/0958-6946(95)00008-9).
- Michalski MC, Briard V, Michel F. Optical parameters of milk fat globules for laser light scattering measurements. *Lait*. 2001;81(6):787–96.
- Michalski MC, Michel F, Geneste C. Appearance of submicronic particles in the milk fat globule size distribution upon mechanical treatments. *Lait*. 2002;82(2):193–208. doi:[10.1051/Lait:2002004](https://doi.org/10.1051/Lait:2002004).
- Michalski MC, Ollivon M, Briard V, Leconte N, Lopez C. Native fat globules of different sizes selected from raw milk: thermal and structural behavior. *Chem Phys Lipids*. 2004;132(2):247–61. doi:[10.1016/j.chemphyslip.2004.08.007](https://doi.org/10.1016/j.chemphyslip.2004.08.007).
- Pal R. Effect of droplet size on the rheology of emulsions. *AIChE J*. 1996;42(11):3181–90.

- Precht D. Fat crystal structure in cream and butter. In: Garti N, Sato K, editors. *Crystallization and polymorphism of fats and fatty acids*. New York: Marcel Dekker; 1988. p. 305–61.
- Tadros T, Izquierdo R, Esquena J, Solans C. Formation and stability of nano-emulsions. *Adv Colloid Interface Sci*. 2004;108:303–18. doi:[10.1016/j.cis.2003.10.023](https://doi.org/10.1016/j.cis.2003.10.023).
- Truong T, Bansal N, Bhandari B. Effect of emulsion droplet size on foaming properties of milk fat emulsions. *Food Bioprocess Technol*. 2014a;7(12):3416–28. doi:[10.1007/s11947-014-1352-4](https://doi.org/10.1007/s11947-014-1352-4).
- Truong T, Bansal N, Sharma R, Palmer M, Bhandari B. Effects of emulsion droplet sizes on the crystallisation of milk fat. *Food Chem*. 2014b;145:725–35. doi:[10.1016/j.foodchem.2013.08.072](https://doi.org/10.1016/j.foodchem.2013.08.072).
- Truong T, Morgan GP, Bansal N, Palmer M, Bhandari B. Crystal structures and morphologies of fractionated milk fat in nanoemulsions. *Food Chem*. 2015;171:157–67. doi:[10.1016/j.foodchem.2014.08.113](https://doi.org/10.1016/j.foodchem.2014.08.113).
- Walstra P. On the crystallization habit in fat globules. *Neth Milk Dairy J*. 1967;21(3/4):166–91.
- Walstra P. Physical chemistry of milk fat globules. In: Fox PF, editor. *Advanced dairy chemistry vol. 2: lipids*. London: Chapman & Hall; 1995. p. 131–78.
- Walstra P, Geurts TJ, Noomen A, Jellama A, Van Boekel MAJS. *Dairy technology: principles of milk properties and processes*. New York: Marcel Dekker, Inc.; 1999.
- Walstra P, Wouters JTM, Geurts TJ. *Dairy science and technology*. 2nd ed. Boca Raton: CRC; 2005.