



## Enhanced characterisation of milk fat globules by their size, shape and refractive index with scanning flow cytometry



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### ABSTRACT

We present a high-precision method for characterisation of a milk sample by the distributions over milk fat globules (MFG) size, shape, and refractive index (RI). We measured light-scattering profiles of individual MFGs and used global optimisation to retrieve their characteristics. We tested two optical models, a sphere and an oblate spheroid, and found that the latter is a more adequate model for part of the MFGs. We applied the developed method to samples of raw bovine milk and milk from two commercial manufacturers. Diameter and RI of individual MFGs were determined with median errors of 74 nm and 0.0094, respectively, which proves the method to be sensitive to small changes in the MFG properties. Moreover, the distributions over size, surface area, and RI showed a good sensitivity to the details of the milk treatment. In particular, the MFG specific surface area is significantly different for all three milk samples studied.

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## 1. Introduction

Since 1674, when Van Leeuwenhoek observed milk fat globules (MFGs) using primitive microscopy, the physical and colloidal properties of these globules have been investigated with different physical methods. This interest has been caused by a role that milk fat plays in health and diseases (Berner, 1993; Spitsberg, 2005). At present, this role is related to different areas of human health, ranging from the obesity problem (Berton et al., 2012) to clearance of apoptotic cells (Lauber et al., 2013). Additionally, the MFGs are responsible for, or contribute to, some of the properties and phenomena observed in liquid dairy products, and are essential to the manufacturing and characteristics of many dairy products (Huppertz & Kelly, 2006).

Surface area can be considered the most important characteristic of the globules, because they are surrounded by a membrane composed of bioactive molecules like proteins, phospholipids,

triglycerides and enzymes (Freudenstein et al., 1979; Lopez, 2011; Singh, 2006). Proteins of the globule membrane interact with milk proteins (Su & Everett, 2003) and microflora (Beresford, Fitzsimons, Brennan, & Cogan, 2001), forming natural bioactive nutrition. The globule membrane is sensitive to modification during isolation and processing, and care should be taken to standardise the composition and characteristics of the membrane to maintain its unique properties during application in food products (Dewettinck et al., 2008). Homogenisation, different types of pasteurisation, sterilisation, etc., can substantially change the surface area of the MFGs of milk samples (Fauquant, Briard, Leconte, & Michalski, 2005). Homogenisation exerts the most dramatic effect on the globules changing their size distribution (Ong, Dagastine, Kentish, & Gras, 2010; Thiebaud, Dumay, Picart, Guiraud, & Cheftel, 2003) and surface proteins (Zamora, Ferragut, Guamis, & Trujillo, 2012).

The mechanisms leading to a particular size distribution of MFGs and changes in their mean size are not well-documented and are still under investigations. The question, “What is the role played by the size distribution of MFGs?” (Lopez, 2011) forces us to develop methods for measurement of this distribution with

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