

**Artificial lipid droplets as a model lipid system**

V. Vezočnik<sup>1</sup>, S. Sitar<sup>2</sup>, K. Kogej<sup>1</sup>, M. Tušek-Žnidarič<sup>3</sup>, V. Hodnik<sup>1</sup>, K. Sepčić<sup>1</sup>, E. Žagar<sup>2</sup>, P. Maček<sup>1\*</sup>

<sup>1</sup>University of Ljubljana, <sup>2</sup>National Institute of Chemistry Slovenia, <sup>3</sup>National Institute of Biology Slovenia

After a century of being underappreciated, natural lipid droplets (LDs) have been finally recognised as important intracellular organelles. Due to the recent discovery of their involvement in various diseases, such as cancer, atherosclerosis or obesity, this area is the subject of intensive research that requires the use of fundamental knowledge. Since the natural systems are complex and unstable, we prepared model artificial LDs with controlled characteristics, using natural lipids.

Our research is aimed to improve preparation and characterization of artificial lipid droplets, nanoemulsions of LDs composed of trioleoylglycerol core covered by a monolayer of sphingomyelin (SM) and cholesterol (Chol). So far, it has been confirmed that those artificial lipid species share many physico-chemical characteristic in common with natural LDs. Furthermore, nanoemulsions covered by a SM/Chol monolayer have been very poorly characterized in contrast to respective SM/Chol vesicles. For that reason, a first detailed characterization of this lipid system has been made using several (bio)chemical and (bio)physical methods and techniques. In parallel, large unilamellar vesicles (LUVs) have been studied, too.

Additionally, a combined modified reverse-phase evaporation/ultrasonication method has been developed for the LDs preparation. Stable LDs and LUVs in controllable sizes in the range of 140 nm to 200 nm for the SM/Chol molar ratio of 1/1 and 4/1 (mol/mol) were prepared and characterized. These LDs and LUVs with defined composition and physical properties were used in studies of interactions of Chol-binding protein perfringolysin O (PFO) as compared to the Langmuir monolayer technique. We suggest that artificial LDs may serve as a useful model lipid system for studying protein-lipid interactions, complementary to the Langmuir monolayers.