

Aim of the study:

The proposed project aims to develop a method of obtaining liposomes with phytosterols or their esters to study their physicochemical and biological properties, as well as thermo-oxidative stability.

Reasons for undertaking the study:

The search for new health-promoting products constitutes one of the recent trends in modern food technology. Unfortunately, many bioactive compounds are perishable and degrade during processing and storage. Another problem is their ability to be absorbed in the human digestive system. Preservation of their biological activity, stability and increased absorption can be achieved by encapsulating such compounds inside liposomes – vesicular structures measuring a few micrometres, which serve as capsules for bioactive substances. An example of such compounds are plant sterols, also known as phytosterols, which have functional properties, yet are easily degradable.

Plant sterols are regarded as the antagonists of cholesterol since they reduce its levels in the human body. They occur naturally in vegetable oils, vegetables and fruit seeds and are added to foods as functional ingredients. Unfortunately, the human body absorbs only about 5% of supplied phytosterols. Furthermore, during food production and storage, these compounds are oxidised, forming derivatives that have adverse effects on the organism and exhibit cytotoxic properties resulting in atherogenic changes and contributing to oxidative stress.

The use of liposomes as carriers of phytosterols should protect them from degradation during thermal-oxidative treatments and increase their absorption in the human body. This will reduce the dose of phytosterols required to lower blood cholesterol levels, which, in turn, will limit the intake of their oxidised derivatives.

Research on the development of new stable structures protecting phytosterols and the impact of their degradation products on the gastrointestinal tract is conducted in the laboratories of the Poznan University of Life Sciences in cooperation with Poznan University of Medical Sciences and Wrocław University of Environmental and Life Sciences. In the age of widely available foods enriched with plant sterols, it is important to ensure that phytosterols are supplied in a form that is completely safe for the consumer. Since it is now known that both free sterols and their esters undergo oxidation to form oxyphytosterols and other derivatives, the development and testing of a new structure, which will undoubtedly be their encapsulation in liposomes, may allow for the creation of a safe and easily absorbed form of these bioactive substances.

Research method:

1. In the first stage of research, a method will be developed to produce liposomes containing free phytosterols or their esters with fatty acids. The structures of the obtained compounds will be confirmed using fluorescence, chromatography, FTIR, DSC or TEM methods.
2. The resulting liposomes will be subjected to heating in the presence of oxygen at 60°C and 180°C, which correspond to the storage test and fat frying, respectively.
3. Phytosterol degradation products will be quantitatively and qualitatively marked using chromatographic techniques, such as GC-FID, GC-MS, GCxGC-MS, HPLC-SEC/ELSD and HPLC-MS in comparison with the corresponding compounds in free or esterified form.
4. Thermal oxidation products of phytosterols and their esters will be fractionated using SPE and HPLC techniques according to their polarity and molecular size into low molecular weight compounds, monomers, dimers and oligomers. Among the thermal oxidation products, the fraction containing oxidised derivatives will also be isolated.
5. This will be followed by *in vitro* studies evaluating the cytotoxicity, genotoxicity and membrane permeability before and after thermal oxidation. Cytotoxicity analyses will be performed on human cells isolated from the small intestinal epithelium, mucosa of the large intestine, liver and vascular endothelium.
6. In the next stage of the project, the liposomes obtained will be subjected to *in vitro* digestion to study their metabolism in the gastrointestinal tract. Their content as well as the level of phytosterols and their oxidised derivatives will be determined in the gastrointestinal epithelial cells.
7. In the last stage, an attempt will be made to implement the newly obtained compounds into food fats and to study their effect on its quality during storage and processing.

The development of the technology of production of liposomes with phytosterols or their esters will be important for the health of consumers because it will not only increase the absorption of phytosterols in the human body, making them more effective in lowering cholesterol levels in human blood, but it will also reduce the consumption of adverse compounds, such as oxyphytosterols.