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ATR-FTIR spectroscopy of extracellular vesicles derived from endothelial cells cultured in hyperglycemic conditions

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Nowadays extracellular vesicles (EVs) are being actively researched. EVs are involved with several biological processes including cell signalling, transfer specific cargo (lipids, proteins, and nucleic acids) and biomarkers of disease. EVs can be divided according to their size and way of arising into exosomes (diameter from 30 nm to 100 nm) and ectosomes (diameter from 100 nm to 1000 nm) [1,2,3].

The Attenuated Total Reflectance Fourier Transform Infrared spectroscopy (ATR-FTIR) method is based on the characteristic absorption of infrared radiation at specific wavelengths by functional groups like N–H, C=O, CH2, CH3, and PO2. An IR spectrum carries specific information on the sample's molecular composition and structure [6,7,8]. The aim of this study is investigation endothelial EVs cargo modifications in hyperglycemic conditions.

In this experiment we used cells, exosomes and ectosomes derived from telomerase-immortalized human microvascular endothelium cell line (TIME) cultured in normoglycemic and hyperglycemic conditions. The parameters were determined to characterize the chemical state of the lipids and proteins of the EVs: saturated to unsaturated fat ratio, acyl chain length, protein phosphorylation and lipid to protein ratio [2,6]. In addition, the percentage contribution of the following secondary protein structures was calculated based on the analysis of the second derivative of the spectra in the Amide I band range: side chain, inter β-sheet, β-sheet, random coil, α-helix and β-turn [2].

FTIR results showed that exosomes, ectosomes and cells differ in content of protein and lipid components. Moreover, obtained results revealed differences in the molecular composition and secondary structures of proteins from EV subpopulations derived from hyperglycemic endothelial cells. Statistically significant differences were found between ectosomes from normoglycemia and hyperglycemia conditions for the values of almost all calculated parameters. Summarizing, ectosomes can be considered as diabetes biomarkers. ATR-FTIR analyses may be useful in identifying new biomarkers of diabetes and its complications.

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