

Evaluation of Cancer-derived Exosomes by FT-IR Spectroscopy

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Exosomes are small nano-sized (30–100 nm) membrane vesicles of endosomal origin released in the extracellular space by a variety of cell types including epithelial cells, mesenchymal stem cells, immune cells and various cancer cells, and are found in all human body fluids. Cancer cells abnormally secrete large quantities of exosomes that transport onco-proteins and immune suppressive molecules thus promoting the growth of tumour and metastasis. Exosomes are composed of proteins, lipids - phospholipids, cholesterol, sphingolipids, ceramides and raft molecules, carbohydrates - mannose, poly lactosamine, α -2,6 sialic acid and complex N-glycan's, which are important to bind proteins in exosome, and nucleic acids such as miRNA, small non-coding RNA and mRNA derived from their cellular origin [1, 2]. In our study FT-IR spectroscopy was used to estimate the purity of extracted exosomes and to evaluate the composition of exosomes produced by colorectal cancer cell lines derived from a primary (SW480) and metastatic (SW620) tumour, which were cultured under hypoxic or normoxic conditions. FTIR spectra were collected using microplate reader HTS-XT (Bruker, Germany). Certainly it was easy to assess the step-by-step purification, to discriminate the fractions of proteins and exosomes and hereafter control the purity of extracted exosomes by FT-IR spectra analyses. In the fraction of purified exosomes the main absorption bands identified proteins ($1550\text{-}1660\text{ cm}^{-1}$), lipids ($2880\text{-}2950\text{ cm}^{-1}$) and carbohydrates ($925\text{-}1111\text{ cm}^{-1}$). Yet the content of nucleic acids was too low for precise detection. The composition of exosomes released by SW480 and SW620 cells significantly differed by the content of proteins. The shift in carbohydrate absorption maximum from 1090 cm^{-1} in normoxic to 1100 cm^{-1} in hypoxic conditions was observed for both exosome samples. It was shown that interrelations of the protein, lipid and carbohydrate content in exosomes varies depending on their origin and is affected by hypoxia.

References

- [1] A.V. Vlassov, S. Magdaleno, R. Setterquist, *Conrad Biochim Biophys Acta* 1820, 940-948 (2012).
- [2] Y. Liang, W.S. Eng, D.R. Colquhoun, R.R. Dinglasan, D.R. Graham, L.K. Mahal, *J Biol Chem.* 289, 32526-32537 (2014).

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