

Abstract

Objectives: This thesis was aimed at studying an albumin transport in vitro using a series of accumulative cell studies with two types of albumin - albumin labelled with the mentioned radioisotope – technetium-99m and fluorescent labelled FITC-albumin. Our task was to verify appropriate procedure of albumin labelling and radiochemical purity. The tasks were also to compare accumulation of FITC-albumin and ^{99m}Tc -albumin in kidney cells, analyze the impact of selected factors on albumin accumulation and determine albumin transmembrane transport mechanisms. Next objective was to cast the results in the form of a scientific statement.

Methods: A low pH, high concentration of albumin and optimum ratio of Sn^{2+} ions. The radiochemical purity of labelled albumin was verified by paper chromatography. Isolated kidney rat cells and standard line of kidney tubular pig cells were used as cell in vitro models. Effects of time, temperature, composition of incubation medium and transport inhibitors were studied in accumulation experiments. Connection between age of cells and albumin accumulation rate was studied in LLC-PK1 cells experiments.

Results: Very pure labelled ^{99m}Tc -albumin was prepared with decreasing radiochemical stability after a few hours. The accumulation of albumin by kidney cells was directly proportional to the number of cells and the amount of albumin. Accumulation parameters of ^{99m}Tc -albumin and FITC-albumin were similar in rat kidney cells experiments. Albumin accumulation rate in young LLC-PK1 cells is compared to older cells severalfold lower. Approximately 3× higher uptake of ^{99m}Tc -albumin was observed during rat cells experiments compared to pig cells. Accumulations of both albumin types were decreased to 20-25 % after incubation at 2°C in kidney cells compared to 37°C incubation.

Conclusions: FITC-albumin and ^{99m}Tc -albumin accumulation studies results are almost equal and both albumin types are acceptable for albumin transport study in vitro. Because of transport interference it is important to look at cells age and type of selected model in cell-line experiments. Active transport mechanisms of young cells do not appear to be appropriately developed. Native cells may have more active protein transport system. The results confirm that large component of albumin uptake in kidney cells has an active character