Time-course analysis of the changes in intracellular amino acid concentrations and amino acid-related signaling pathways in cancer cells induced by the inhibition of cancer-type amino acid transporter LAT1

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There is an increased demand for nutrients in cancer cells compared to normal cells. Accordingly, the nutrient intake and metabolism in cancer cells are regarded as promising therapeutic targets. L-type amino acid transporter 1 (LAT1) is upregulated in cancer cells of various tissue origin, and has a large effect on the uptake of amino acids. LAT1 inhibitors have been shown to suppress cell proliferation and tumor growth *in vitro* and *in vivo*. However, it has not been clarified yet how the intracellular concentration of each amino acid changes and, concomitantly, what cellular responses are induced in cancer cells when treated with LAT1 inhibitors. In this study, we established a high performance liquid chromatography (HPLC) system to quantify amino acids. Using the system, we could successfully follow the time course of the change in the concentration of cellular amino acids in human pancreatic cancer MIA PaCa-2 cells treated with LAT1 inhibitor. In the same experimental condition, we also detected changes in the amino acid-related signaling pathways including mTORC1 pathway and autophagy. Currently, we are monitoring the changes in the amino acid concentrations in the cell culture medium. The detailed effects of LAT1 inhibitors on the intra- and extracellular amino acids, and the subsequent cellular responses will be discussed.