CTOS as a model for studying tumor heterogeneity and plasticity

Masahiro Inoue, Jumpei Kondo
Department of Clinical Bio-resource Research and Development
Graduate School of Medicine Kyoto University

Cancer is a disease characterized by its heterogeneity and plasticity. Cancer patients with the same pathological diagnosis differently respond to the therapy, and cancer cells in a tumor are genetically and phenotypically divergent. On the other hand, cancer cells change their characters, such as metastatic potential and drug resistance, during the clinical course. Such heterogeneity and plasticity make the cancer treatment difficult. Interpatient heterogeneity is a critical issue in molecular targeting drugs because they are very well effective on some patients, but not effective at all on the others. Since the effects can be predicted by assessing the mutations of the genes related to the targeted pathway in some cases, genome sequencing is going to be widely applied in clinic. Since it is realistically difficult to predict the optimal drug for majority of the patients only by mutational analysis, additional approaches are required to make the prediction better. Sensitivity assays using primary cultured cells has long been expected to be a prediction platform but none of them are widely used so far.

We developed a novel primary culture system of cancer cells, cancer tissue-originated spheroid (CTOS) method, in which cancer cells are prepared and cultured by maintaining cell-cell contact throughout the process. CTOS preserves the nature of cancer cells in original patient tumors. Some of the CTOS can be xenografted, and from the xenograft tumors, CTOS can be easily and efficiently prepared and cryo-preserved. We call the CTOS as 'CTOS lines' when CTOS can be passaged in vivo at least twice and a number of cryo-tubes are stocked for further studies. We generated CTOS panels, which consist of multiple CTOS lines prepared from different patient tumors. Using CTOS panels, we revealed wide range of drug sensitivity among the lines depending on the drug. The sensitivity assay reflects the results of clinical trials as well as the clinical response of individual patients.

As for the plasticity, CTOS dynamically changes their characteristics as a mass, not just a single cell. One of the examples is the polarity switching. Most of the colorectal cancer is the differentiated adenocarcinoma, in which gland like structure, apico-basal polarity as a cell, is maintained in various extents. We previously reported that polarity status of CTOS switched according to different culture conditions; apical-out in suspension and apical-in in extracellular matrix (ECM). Micropapillary carcinoma (MPC) is an aggressive variant of adenocarcinoma, exhibiting reverse polarity, i.e. apical side facing to the ECM. The machinery of MPC has not been clarified yet. We established CTOS lines from MPC type colorectal cancers, MPC-CTOSs. MPC-CTOS formed apical-out pattern even in ECM, recapitulating the polarity status in original tumors. Treatment with ROCK inhibitor Y27632 enabled MPC-CTOS to accomplish polarity switching in ECM. The polarity status of xenograft tumors of C166, one of the MPC-CTOSs, changed to that of non-MPC when Y27362 was administered in vivo. These results suggest that RhoA/ROCK signaling pathway might be involved in MPC phenotype.

Taken together, CTOS can be a model for studying tumor heterogeneity and plasticity.

[References]

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井上 正宏
京都大学大学院医学研究科　クリニカルバイオリソース研究開発講座　特定教授

1987年　大阪大学医学部卒
1987年　臨床研修　大阪大学第一外科、小児外科など
1991年　大学院　大阪大学小児外科、大阪大学細胞工学センター
1998年　博士研究員　UC San Francisco
2001年　大阪国際がんセンター（旧 大阪府立成人病センター）生化学部　部長
2011年　大阪大学大学院薬学研究科　環境病因病態学分野　客員教授
2013年　京都大学大学院医学研究科　客員教授
2015年　大阪大学大学院医学研究科　客員教授
2018年　京都大学大学院医学研究科　クリニカルバイオリソース研究開発講座　特定教授