Evaluation of Cell-able Oncology™ spheroid culture system for culturing patient derived primary tumor cells

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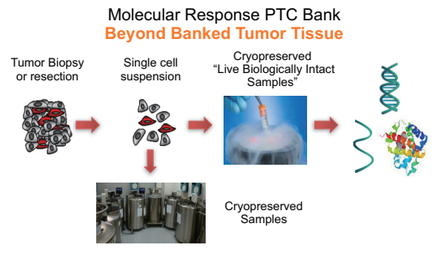
Abstract

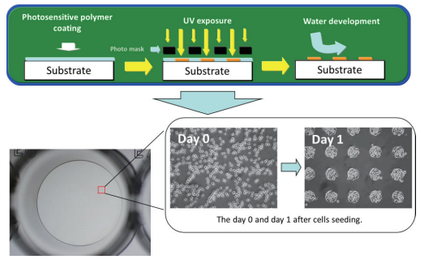
Tumor cell microenvironment has significant impact on growth kinetics, cell signaling and response to drug treatments. 3D models are more biologically relevant models compared to 2D models and have gained preference among researchers and drug developers. It is well recognized that primary tumor cell cultures grown in 2D monolayer quickly lose the ability to proliferate while 3D culture conditions on extracellular matrix allows for spheroid formation and proliferation. Cell-able OncologyTM, a novel micropatterned plate has cell adhesion areas in the well bottom where the size and shape of these areas are tightly controlled. Micro-fabrication of the well bottom enables rapid cell attachment and spheroid formation. Cell-able OncologyTM plates are potentially an alternative to culturing cells in 3D without extracellular matrix. We have evaluated spheroid formation, morphology, duration of viability, histopathology and expression of markers in patient-derived primary tumor cells from different tumor types when cultured on Cell-able OncologyTM plates compared to Cultrex extracellular matrix (ECM) coated plates. In addition we have utilized Cell-able plates to grow patient-derived tumor cells and evaluate the anti-proliferative responses to cytotoxic and targeted agents using high content imaging coupled with immunofluorescence characterization enabling subpopulation analysis within the spheroids. Our results indicate that patient-derived tumor cells from multiple indications as well as tumor cell lines can be cultured on Cell-able OncologyTM plates as spheroids that resemble the micro architecture of tumors and are therefore a suitable 3D model. The results of high content imaging analysis suggest that Cell-able OncologyTM plates could be used as a valuable tool in in vitro predictive assay models in oncology drug development.

Patient Derived Primary Tumor Cells



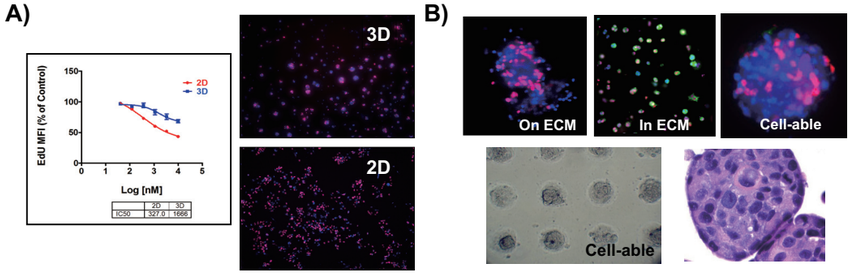
Creation of companion Biobank: Matched DNA, RNA and protein from cryopreserved cells
- DNA: mutations, copy number, translocation
- RNA: gene expression, splice variation
- Protein: antigen detection, size variation

Cell-able Oncology™ Spheroid culture system



Photolithographic process to fabricate 3-dimensional cell structure: Chemical surface modification using photosensitive & non cell-attaching polymer on the cell culture plate.

High Content Imaging in Patient Derived Primary Tumor Cells in 3D environment

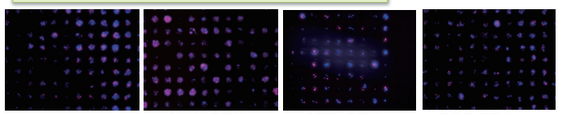


***Ex vivo* studies on Tumor cells: 2D vs 3D: (A)** Comparison of 3D and 2D assays: Action and potency of cancer drugs depend on the culture conditions of tumor cells *ex vivo*. (**B)** Patient-derived tumor cells can be grown as tumor spheroids using different methods. Blue: DAPI, Green: Cytokeratin, Red: EdU

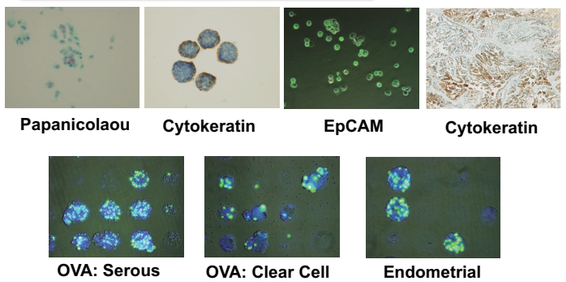
Cell-able Spheroid Culture System: A Powerful Tool for Oncology Drug Discovery Programs

Cancer Cells Grow as Spheroids on Cell-able Oncology™ Plates

Patient-derived tumor samples from bio-bank

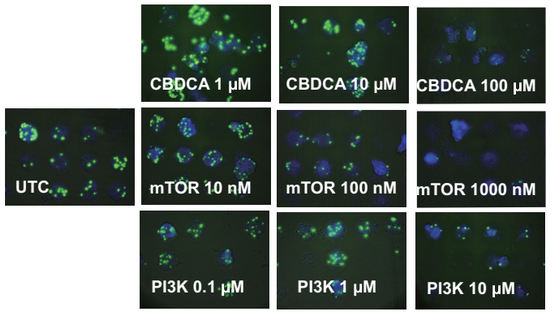


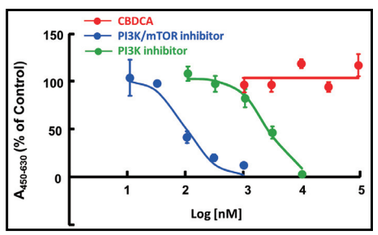
Fresh (Non-cryopreserved)Tumor samples



Cell-able Oncology™ plates as a novel 3D spheroid culture: Patient-derived tumor cells (freshly collected as well as cryopreserved) grow as 3D spheroids on Cell-able OncologyTM plates. Tumor cell lines also form spheroids on Cell-able OncologyTM plates.

Cell-able Oncology™ plates as a tool for drug response studies

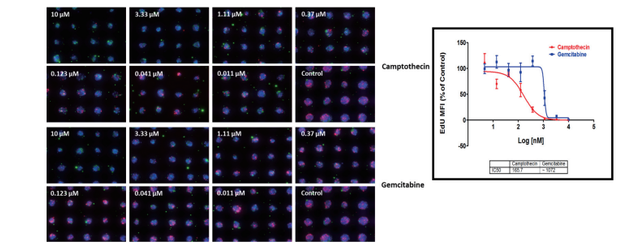


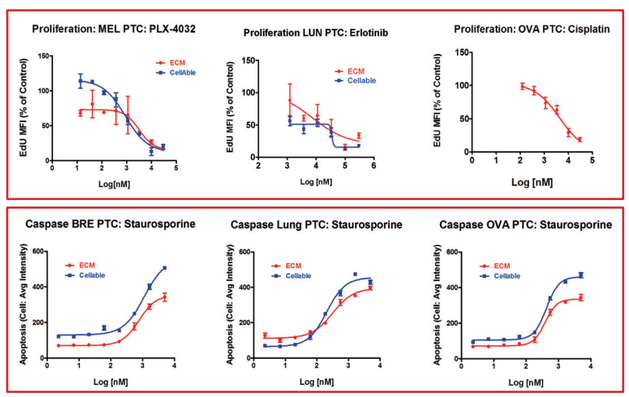


	CBDCA	PI3K/mTOR inhibitor	PI3K inhibitor
IC50	>100000	85	3000

Use of Cell-able Oncology™ plates for ex vivo treatment studies: Patient-derived tumor cells (freshly collected) were grown as 3D spheroids on Cell-able OncologyTM plates and were treated with oncology drug candidates. Change in the proliferation were visualized by EdU uptake and quantified using WST-8 Assay. Blue: DAPI, Green: EdU

High-Content Imaging of Tumor Spheroids in Cell-able Oncology™ Plates



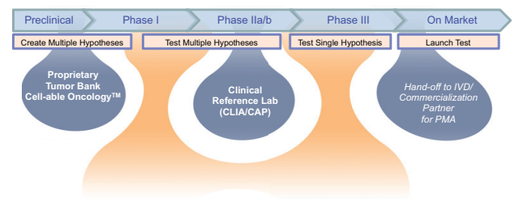


Combining Cell-able Oncology™ plates with High-Content Imaging Platform: Patient-derived tumor cells (cryopreserved) were grown as 3D spheroids on Cell-able OncologyTM plates and were treated with oncology drug candidates. Change in the proliferation were visualized by EdU uptake, apoptosis by Caspase 3/7 assay. Imaging and analysis were performed using ImageXpress Micro and MetaXpress (Molecular Devices, Sunnyvale, CA). Blue: DAPI, Red: EdU

Novel 3D culture system for Oncology Drug Discovery

- ◆Patient-derived tumor cells and tumor cell lines grow into spheroids.
- ◆Compatible with relevant endpoints used in drug discovery.
- ◆Easy to handle and image compared with 3D ECM culture.
- ◆Similar drug sensitivity to 3D ECM culture.
- ◆Applicable to use in basic through clinical research in oncology.

Integrated Approaches to Discovery and Clinical Utility



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