

Evaluation of anticancer agents using patient-derived tumor organoids that are maintained the characteristic of tumor tissues

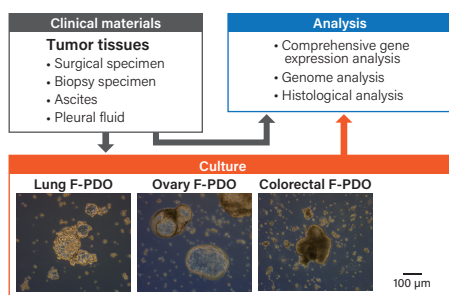
Hirosumi Tamura, Arisa Higa, Hiroataka Hoshi, Gen Hiyama, Nobuhiko Takahashi, Jun-ichi Imai, Shinya Watanabe, Motoki Takagi
 Medical-Industrial Translational Research Center, Fukushima Medical University

Abstract

Fukushima patient-derived tumor organoids (F-PDOs), which can be cultured for over a period of 6 months and form cell clusters with similar morphologies to their source tumors, were established from human tumor tissues, including those from the lungs, ovaries, uterus, and so on. The comparative histological, genome, and comprehensive gene expression analyses between F-PDOs and their source tumor tissues proved that the characteristics of F-PDOs were similar to those of their source tumors. At present, 79 F-PDOs have been established. Herein, we report the detailed analysis of F-PDOs established from the endometrial tumor tissues from these F-PDOs.

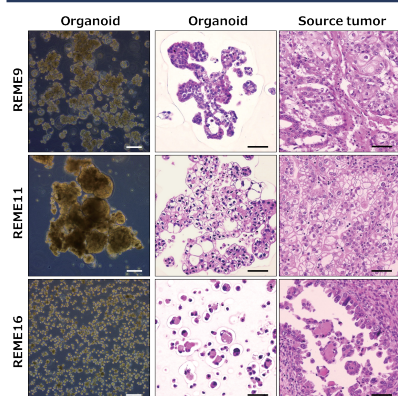
F-PDOs (REME9, 11, and 16) established from endometrial tumor tissues were used for experiments on the cell growth inhibition of anticancer agents. A suitable high-throughput assay system, with 96- or 384-well plates, was designed for each F-PDO, and the anticancer agents were evaluated. REME9 and 11 exhibited distinct responses and increased resistance to the drugs, as compared with conventional cancer cell lines. REME9 and 11, which were established from tumors that originated in patients who did not respond to paclitaxel and carboplatin (the standard chemotherapy for endometrial cancer), exhibited high resistance to the two agents. Therefore, assay systems using F-PDOs may be utilized to evaluate anticancer agents using conditions that better reflect clinical conditions, compared with conventional methods using cancer cell lines, and to discover markers that identify the pharmacological effects of anticancer agents.

Establishment of F-PDOs



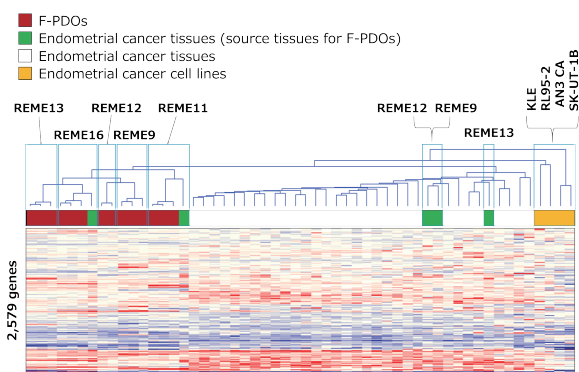
- ✓ F-PDOs were defined to possess the following properties: they were able to be cultured for >6 months period and their characteristics are highly similar to those of their source tumors determined by comprehensive gene expression analysis, genome analysis, and histological analysis.
- ✓ 79 F-PDOs were established from human lung, breast, ovarian and uterine tumor tissues, among others.

Histological analysis of F-PDOs



- ✓ The morphology of F-PDOs were similar to the source tissues.

Gene expression profiles of F-PDOs



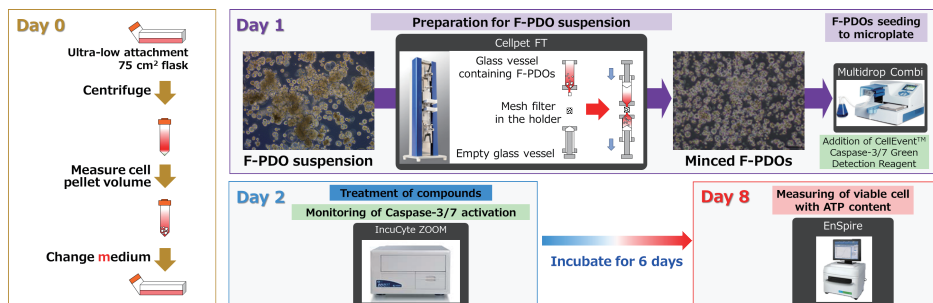
- ✓ The gene expression of F-PDOs possessed characteristics of the source tissues, although they were not similar to endometrial cell lines.

Unsuitable characteristics of F-PDOs for assay

- Cell growth is slow.
 - Difficult to obtain required cell amount
- Size is various and non-uniform.
 - Difficult to seed equally on each well.
- Morphology is a large cluster.
 - Impossible to measure cell number.
- Maintenance of culture condition is difficult.
 - More experience is necessary.

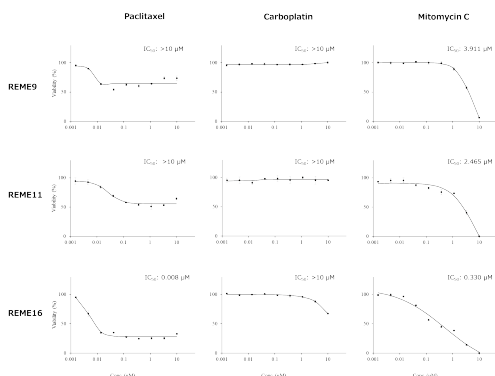
"Know-how" is indispensable for assays that involve F-PDOs.

Cell growth inhibition assay of anticancer agents using F-PDOs in multi-well plates



Growth inhibition assay of anticancer agents using F-PDOs in 96-well plates

Dose-response curve of anticancer agents in F-PDOs

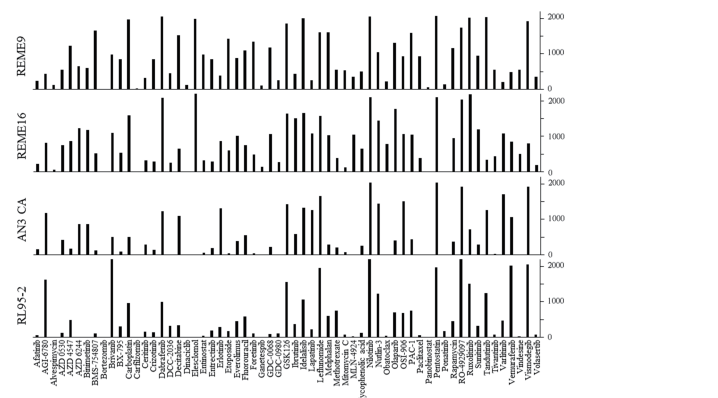


- ✓ REME9 and 11 lines were more resistant to all drugs than REME16.
- ✓ REME9 and 11 lines have high resistance to paclitaxel and carboplatin, which were used for standard chemotherapy for endometrial cancer.

High-throughput assay of anticancer agents using F-PDOs in 384-well plates

Area under the curve (AUC)

The AUC values were in the range of 0.1–20 μM.



- ✓ The drug responses of REME9, and 16 were varied.
- ✓ The overall sensitivity of REME9 and 16 to anticancer agents was lower compared with that of the cell lines.

Conclusion

- ◆ Comparative histological and comprehensive gene expression analyses proved that the characteristics of PDOs were similar to those of their source tumors.
- ◆ CellPet FT, which is able to be dispensed equally into each well by mincing F-PDOs, enabled a high-throughput assay system for the evaluation of anticancer agents.
- ◆ The assay system using F-PDOs may facilitate the evaluation of anticancer agents under conditions that reflect clinical conditions more accurately compared with conventional methods.
- ◆ F-PDOs may aid the discovery of new candidates of drugs and markers to predict the pharmacological effects of anticancer agents.