Project title: Translational Research for Improving Cancer Patient Survival in Taiwan-A Multiple Center Approach Program title: Colorectal cancer Culture of Isolated Circulation Tumor Cell in Patients with Colorectal Cancer-Preliminary Result

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Introduction: Presence of circulating tumor cells (CTCs) is related to develop subsequent distant metastasis of colorectal cancer. In this study, we establish the in vitro cultivation of CTCs from patients with colorectal cancer, and evaluate its relation to tumor characteristic.

Patients and methods: CTCs isolated from peripheral blood of CRC patients by microfluidic platform CMx was carried out, then primary CTC culture with specific indicated medium was done. The immunocytochemistry staining of cultured cells included the marker of CK20 (Abcam) for CRC cancer cell, a-SMA (a-smooth muscle actin, DAKO) and FGFR (fibroblast growth factor receptor, Abcam) for fibroblast staining, CD45-FITC (DAKO) for leukocyte staining, CellTracker Red CMTPX (Invitrogen) used as cell stain for metabolic cells during cell culture; and DAPI or Hoechst33342 used for routine nuclear staining. The single isolated, clustered CTCs, cultured single and colonized CTCs were calculated and their relation to tumor characteristic were analyzed.

Results: The successful culture rate was 90% (36/40 patients). We found the colony number of cultured CTC was significantly increased in patients with metastasis (p=0.039). The number of single cultured CTC was significantly high in patients with CEA elevation as compared to those not (P=0.011). The number of isolated single CTC and cluster CTC is related to pT classification and tumor size > 5 cm in diameter, but not related to pN classification.

Conclusion: The viability of isolated CTCs from the patients were good with high successful culture rate. High isolated and culture CTC number is associated with distant metastasis, tumor invasion depth of bowel and tumor size.