Clinical Research (Excluding Clinical Trials)

Abstract 489: NGS analysis of the single CTC or DTC isolated and subtyped by the integrated subtraction enrichment and immunostaining-FISH

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Abstract

Majority of the circulating or disseminated tumor cell (CTC or DTC) detection methodologies rely on existence of epithelial markers (such as EpCAM and/or cytokeratins) on tumor cells. However, highly heterogeneous expression or even absence of EpCAM, and down-regulation of both EpCAM and cytokeratins in CTCs/DTCs during epithelial-mesenchymal transition (EMT) has been reported elsewhere, resulting in significant false negative detection of those uncatchable and invisible CTCs/DTCs. A novel strategy integrating subtraction enrichment which efficiently enriches non-hematopoietic tumor cells shed from various types of solid tumor into biofluid including peripheral blood, bone marrow, ascites, pleural effusion, and cerebrospinal fluid (CSF), with immunostaining-FISH (iFISH), which enables in situ phenotypic immunofluorescent staining of tumor biomarker and chromosomal FISH on the identical tumor cell, has shown its unique advantage in terms of detecting CTCs/DTCs with higher sensitivity and specificity. Diversified CTCs/DTCs could be identified and subtyped upon specific chromosome ploidy and tumor biomarker expression, and each subtype of CTCs/DTCs respectively correlate with therapeutic drug sensitivity or resistance, tumor metastasis or relapse. Whole genome amplification and subsequent next generation sequencing (NGS) analysis performed on the single subtyped breast, pancreatic, gastric and NSCLC cancer CTC or DTC targeted by iFISH, and collected by means of a microscopic cell manipulator is able to reveal ploidy of all the chromosomes and status of either unknown genes or a series of known oncogenes including HER2, Kras, BRAF, etc. It is anticipated that SE-iFISH could guide and promote more significant single cell based genomic and functional analyses of CTCs and/or DTCs.

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