**Circulating Tumour Cells as Liquid Biopsies in Clinical Studies: the example of TRACERx**

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Tracking Cancer Evolution through Treatment (TRACERx) is an ongoing Cancer Research UK funded prospective study of the evolution of Stage I-IIIA NSCLC currently being undertaken across a UK wide consortium led from the CRUK Lung Cancer Centre of Excellence by Charles Swanton. Patients undergo surgical resection of their disease with curative intent. Tumours are spatially separated and comprehensively profiled to generate phylogenetic trees; of note a high proportion of subclonal copy number alterations (CNA) were associated with recurrence and death (Jamal-Hanjani et al, NEJM, 2017). Patients who relapse undergo a biopsy of their metastatic disease. Liquid Biopsies (CTCs and ctDNA) are performed periodically from prior to resection through to relapse and death and patients may consent to the CRUK funded PEACE post mortem study. TRACERx therefore studies NSCLC progression and metastasis from diagnosis to death and our hypothesis is that analysis of extensive tissue and liquid biopsies will help distinguish metastatic competent from incompetent subclones in the same patient. We enumerated CellSearch CTCs in the pulmonary vein draining the cancerous lung (PV-CS-CTCs) just prior to tumour resection in 122 patients and asked whether the presence of CTCs could predict relapse. Where sufficient CTCs were detected, we isolated pools or single CTCs and examined CNA and mutations and compared CTC profiles with those of resected tumour and metastatic biopsies. In this presentation I will present our new data describing a) our robust workflow for enrichment, enumeration, isolation, banking and molecular analysis of single CTCs is established, b) single cell whole genome analysis and next generation sequencing applied to PV-CS-CTCs from 16 early stage NSCLC patients ~ > 100 CTCs and white blood cells, c) CTC heterogeneity and presence of PV-CS-CTCs with overlapping genetic changes in common with the resected tumour regions, d) PV-CS-CTCs (by morphology) that do not share CNA with the corresponding tumour and e) that in the one patient case studied in depth, PV-CS-CTCs share a common progenitor with the metastasis and are more representative of relapse than the bulk primary tumour. These exciting early case report data imply that PV-CS-CTCs harbour mutations not seen in the primary tumour yet detected in the metastasis presenting 11 months post-surgery and serve as a paradigm of the power of CTC analysis to identify lethal subclones in NSCLC.