

Land of hope and dreams

Selection of life science and translational medicine literature

by Marco Confalonieri

The diagnostics of cancer is rapidly changing. To date biopsies are still the core of any final oncologic diagnosis. Nevertheless, the molecular characteristics of cancer are becoming more and more important to properly prescribe the newest oncologic therapies. The technological advances of the past decade have helped researchers to characterize the molecular basis of cancer. With this knowledge, drugs targeted at specific molecules have been designed. New rapid molecular diagnostics, or biomarkers, for cancer are now urgently needed to determine which of these new drugs a patient is most likely to benefit from.

The Cancer Genome Atlas Research Network has recently published in *Nature* (1) a comprehensive molecular profiling of lung adenocarcinoma. Findings included a high rate of somatic mutations including alterations in tumor-suppressor genes, chromatin-modifying genes, and RNA-splicing genes and suggested novel or as-yet unidentified mechanisms in pathway activation. These data establish a foundation for classification and further investigations of lung adenocarcinoma molecular pathogenesis. Circulating tumour cells (CTCs) are rare cells that are shed from primary and metastatic tumour deposits into the peripheral circulation. The detection of these cells into the bloodstream represents a means of performing non invasive tumour sampling for prognostic and therapeutic purpose. Indeed, enumeration of CTCs has shown to correlate with cancer stages, but because their number can be very small for apoptosis and death these cells are not easily detected. Fortunately, scientists have discovered that dying tumour cells regularly release small pieces of their DNA into the bloodstream. These pieces are called cell-free circulating tumour DNA (ctDNA). So, another promising technique for the non-invasive diagnosis of cancers is the detection of tumour DNA in the bloodstream. Moreover, circulating tumour DNA (ctDNA) could guide personalized cancer treatment through a timely identification of its molecular characteristics. By developing and refining techniques for measuring and sequencing ctDNA scientists are turning vials of blood into "liquid biopsies" providing more comprehensive information than conventional biopsies do. Recently, investigators from Stanford University using a modified cheap ctDNA technique identified a small proportion of the genome that is repeatedly mutated in lung cancers (2). Diehn et al. used data from over 400 patients to develop a sequencing approach that covers recurrent gene mutations in lung cancer (2). With this method, they detected ctDNA with high specificity in all samples from another set of patients with stage II and advanced disease and half of the samples from stage I cancer patients. The amounts of ctDNA measured correlated with tumour volume during the course of thera-



py, identified patients with residual disease after treatment, and better detected response to therapy compared to radiographic methods.

1) Comprehensive molecular profiling of lung adenocarcinoma

The Cancer Genome Atlas Research Network
Nature 2014;511:543-550

Abstract

Adenocarcinoma of the lung is the leading cause of cancer death worldwide. Here we report molecular profiling of 230 resected lung adenocarcinomas using messenger RNA, microRNA and DNA sequencing integrated with copy number, methylation and proteomic analyses. High rates of somatic mutation were seen (mean 8.9 mutations per megabase). Eighteen genes were statistically significantly mutated, including RIT1 activating mutations and newly described loss-of-function MGA mutations which are mutually exclusive with focal MYC amplification. EGFR mutations were more frequent in female patients, whereas mutations in RBM10 were more common in males. Aberrations in NF1, MET, ERBB2 and RIT1 occurred in 13% of cases and were enriched in samples otherwise lacking an activated oncogene, suggesting a driver role for these events in certain tumours. DNA and mRNA sequence from the same tumour highlighted splicing alterations driven by somatic genomic changes, including exon 14 skipping in MET mRNA in 4% of cases. MAPK and PI(3)K pathway activity, when measured at the protein level, was explained by known mutations in only a fraction of cases, suggesting additional, unexplained mechanisms of pathway activation. These data establish a foundation for classification and further investigations of lung adenocarcinoma molecular pathogenesis.

2) An ultrasensitive method for quantitating circulating tumour DNA with broad patient coverage

Newman AM, Bratman SV, To J, et al.
Nat Med 2014;20:548-554

Abstract

Circulating tumour DNA (ctDNA) is a promising biomarker for non invasive assessment of cancer burden, but existing ctDNA detection methods have insufficient sensitivity or patient coverage for broad clinical applicability.

Here we introduce cancer personalized profiling by deep sequencing (CAPP-Seq), an economical and ultrasensitive method for quantifying ctDNA. We implemented CAPP-Seq for non-small-cell lung cancer (NSCLC) with a design covering multiple classes of somatic alterations that identified mutations in >95% of tumours. We detected ctDNA in 100% of patients with stage II-IV NSCLC and in 50% of patients with stage I, with 96% specificity for mutant allele fractions down to ~0.02%. Levels of

ctDNA were highly correlated with tumour volume and distinguished between residual disease and treatment-related imaging changes, and measurement of ctDNA levels allowed for earlier response assessment than radiographic approaches. Finally, we evaluated biopsy-free tumour screening and genotyping with CAPP-Seq. We envision that CAPP-Seq could be routinely applied clinically to detect and monitor diverse malignancies, thus facilitating personalized cancer therapy.