Functional lung adenocarcinoma risk SNPs identified through positional integration with human alveolar epithelial cell epigenomes

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Motivation: Lung cancer is the leading cause of cancer death, and lung adenocarcinoma (LUAD) is its predominant histological subtype. Fifteen single nucleotide polymorphisms (SNPs) have been significantly associated with LUAD risk in genome-wide association studies (GWAS). However, because most GWAS SNPs reside in non-coding regions and are co-inherited with hundreds of SNPs in linkage disequilibrium (LD), which SNPs play a causal role in disease development remains largely unknown. We hypothesized that some of these SNPs affect oncogenic transcriptional programs by modulating the activity of gene enhancers in alveolar epithelial cells (AECs), the purported cells of origin for LUAD.

Methods: To test this hypothesis, we overlaid epigenomic features of primary human AECs over the locations of index LUAD risk SNPs and associated high LD SNPs. Luciferase assays for enhancer activity were performed for candidate SNPs that were predicted to disrupt transcription factor (TF) binding sites in loci marked by features of active enhancers. Expression quantitative trait loci (eQTL) analysis was also performed using The Cancer Genome Atlas (TCGA) dataset and the online Genotype-Tissue Expression (GTEx) Portal to identify potential target genes of each SNP-enhancer pair.

Results: Thirty-three LUAD risk-associated SNPs mapped to putative AEC enhancer regions. TF binding site prediction suggests that numerous SNPs might alter the binding affinity of TFs implicated in lung cancer, including RXRA and NKX2-1. Luciferase assays indicate that two of the SNPs significantly affect enhancer activity in lung cancer cell lines. eQTL analyses link each of the putative enhancers to candidate target genes, including both known oncogenes and genes not previously associated with lung cancer.

Conclusions: Taken together, our analyses provide new mechanistic insight into long-known associations between non-coding SNPs and LUAD outcomes, and may ultimately yield more effective and personalized strategies for lung cancer risk assessment, prevention, and treatment.

Magnetic nanocubes for selective capture of circulating tumor cells in NSCLC

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Non-small cell lung cancer (NSCLC) is the leading cause for cancer related mortality rates in the US, often associated with 20-35% response rate and a ~10 month median survival time. Currently, tissue diagnostics is performed using immunohistochemistry, FISH, and PCR for staging and treatment planning. In vivo imaging such as PET or CT is also used to detect the severity of NSCLC. NSCLC metastasize by spreading primary tumor cells to distant organs. Therefore, it is possible to isolate circulating tumor cells (CTC) in patients’ blood, and as the cells originate from tumor, detailed genetic evaluation about the tumor could be performed. Isolation and study on circulating tumor cells is slowly evolving as liquid biopsy of cancer. In fact, CTC detection technique is emerging as prognostic markers to identify treatment response in NSCLC patients. The current CTC capture methodology involves cell search technologies, predominantly relying on EpCAM expression based detection. However with the discovery of tumor heterogeneity and consequent impact on clinical treatment, it is important to detect patients with CTCs early on, based on their genetic alterations. Both HER2 (2-5% mutation incidence) and EGFR (10-35% mutation incidence) overexpression have been pronounced in patient biopsies and their exclusivity in individuals has been seen as a prerequisite in chemotherapeutic selection and dose. Moreover there is no standardized process to selectively identify CTCs based on HER2 and EGFR surface expressions. While EGFR and resistant mutations has been prominent, in the understanding of NSCLC characterization, HER2 is relatively less explored and frequently associated with breast cancer detection. Recent studies show that HER2 expressions correlate with metastases and disease free survival. Overall this fact leads us to believe HER2 markers could be present in CTCs. Therefore, we are developing magnetic iron nanocubes (FeNC) functionalized with either Herceptin or Cetuximab as markers. These CTC markers can be correlated with tumor heterogeneity and decide therapeutic targets for first line and second line treatment. Our approach involves cell sensing using magnetic nanoparticles (MNPs), counting and subsequent separation of live A549 (HER2 +ve; EGFR +ve) and HCC827 (HER2 -ve; EGFR +ve) cells from a mixture for further processing. In our current work we have synthesized HER2 and EGFR receptor targeting iron oxide nanoparticles (50nm) that