The druggable mutation landscape of lung cancer

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Abstract

Background: Molecularly targeted therapies and immunotherapies have emerged as promising approaches in the fight against advanced-stage cancers, including lung adenocarcinoma. Identifying genomic alterations predictive of targeted therapy response, as well as biomarkers for immunotherapy response, such as tumor mutation burden (TMB), could minimize the utilization of ineffective therapies and help overcome tumor drug resistance, improving patient outcomes. We examined the largest previously described dataset, consisting of genomic alterations of ~10,000 lung adenocarcinoma patients, to characterize the landscape of druggable alterations and identified previously undetected co-occurrence and exclusivity relationships between genomic alterations.

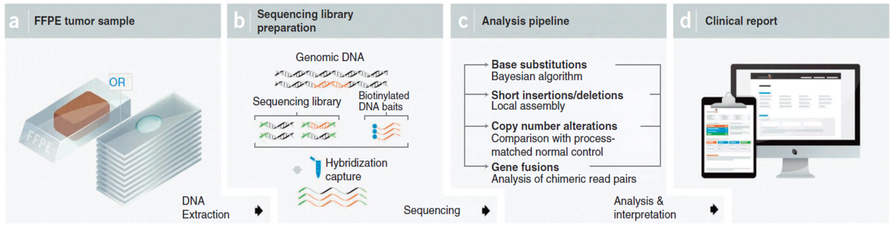
Methods: Comprehensive genomic profiling (CGP) based on hybrid capture-based next-generation sequencing of the full coding regions of up to 315 cancer-related genes was performed on 10,472 lung adenocarcinomas. Base substitutions, indels, copy number alterations and gene fusion/rearrangements were assessed. TMB was calculated as the number of somatic, coding, base substitutions and indels per megabase of genome examined (high TMB: ≥20 mutations/Mb).

Results: Patient median age was 65 years (range: 13 to >90 years), and 56% were female. The alteration frequencies of the druggable NCCN lung adenocarcinoma guideline genes were: *EGFR* (20.5%), *BRAF* (5.8%), *ERBB2* (5.7%), *c-MET* (5.2%), *ALK* (4.3%), *RET* (2.1%), and *ROS1* (1.4%). 57.5% and 2.5% of samples had alterations in zero or multiple of these genes, respectively. Few cases with high TMB were found in samples with alterations in *EGFR* (3.6%) or *ALK* (2.6%), while a larger percentage with alterations in BRAF (12.9%) or zero NCCN genes (17.4%) had high TMB. 269 cancer-related genes were each altered in ≥0.1% of cases without alterations in NCCN genes or high TMB, including genes that are becoming clinically relevant, such as *STK11* (24.8%), *MYC* (8.4%), *NF1* (8.2%), *PIK3CA* (5.2%), *RICTOR* (3.7%), *CDK4* (2.8%), *CCND1* (2.8%), *BRCA2* (1.7%), and *NRAS* (1.3%). Detailed co-occurrence and exclusivity relationships for all genomic alterations will be presented. *EGFR*, *RET*, and *ROS1* alterations were most common in female cases, and *ALK*- and *ROS1*-altered tumors had the lowest patient age distributions (medians: 57 and 55 years, respectively).

Conclusion: Using CGP, >50% of patients with lung adenocarcinoma had an alteration in at least one NCCN gene (42.5%), a high TMB status (12.3%), or both (2.3%). Amongst those with neither, 47.5% had an alteration in a gene with emerging evidence for clinical utility. Given the robustness of the dataset, this analysis suggests an expansion of the patient population eligible for personalized anti-lung cancer treatment through combination therapy and immunotherapy.

Methods

Comprehensive cancer genome profiling assay workflow



Sample requirements
- Surface area: ≥25 mm^2
- Sample volume: ≥1 mm^3
- Nucleated cellularity: ≥80% or ≥30,000 cells
- Tumor content: ≥20%

Fraction of patients with tissue insufficient for analysis: 10–15%

Laboratory process highlights
- Requires ≥50 ng of dsDNA (quantified by PicoGreen)
- Fragmentation by sonication (Covaris) and 'with-bead' library construction
- Hybridization capture with biotinylated DNA oligonucleotides
- 49 × 49 paired-end sequencing on the Illumina HiSeq platform to >500× average unique coverage, with >100× at >99% of exons

Analysis methods highlights
- Sensitivity to variants present at any mutant allele frequency
- Detection of long (1–40 bp) indel variants using de Bruijn graph–based local assembly
- CGH-like analysis of read-depth for CNAs assessment

Reporting approach

Interpretation without a matched normal
- Germline variants from 1000 Genomes Project (dbSNP135) removed
- Known driver alterations (COSMIC v62) highlighted as biologically significant

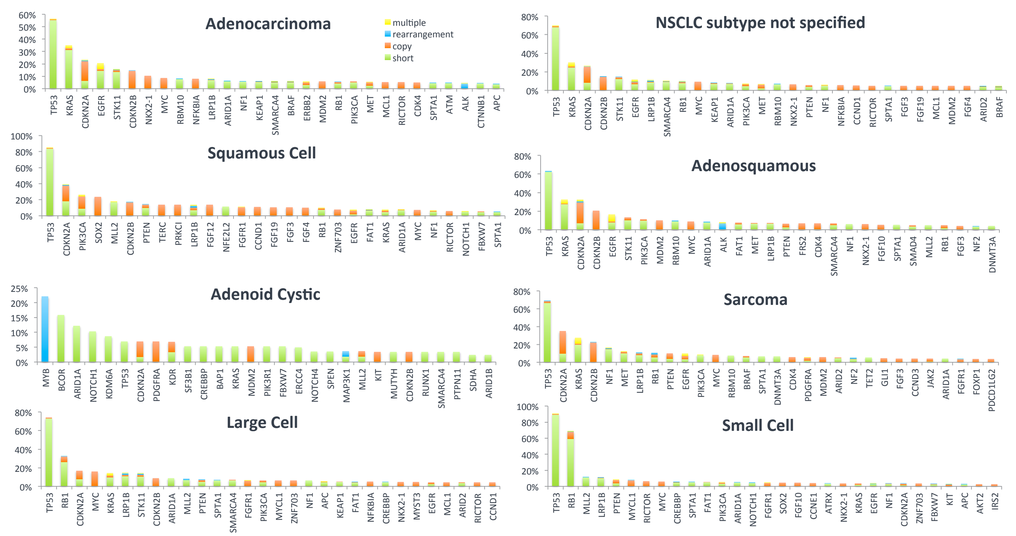
A concise summary of the biomedical literature and current clinical trials is provided for each highlighted alteration

Cohort Demographics

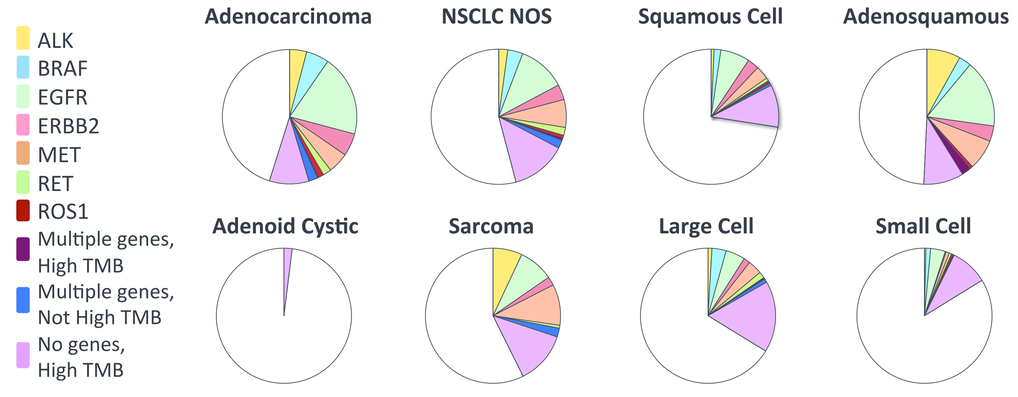
	Adenocarcinoma	NSCLC subtype not specified	Squamous	Adenosquamous	Adenoid Cystic	Sarcoma	Large Cell	Small Cell
Number of Patients	9,794	2,157	1,660	130	51	137	314	878
Percent Male/Female	44% / 56%	52% / 48%	63% / 37%	48% / 52%	43% / 57%	58% / 42%	55% / 45%	49% / 51%
Age: 5th Percentile	44	44	48	46	29	46	40	43
Age: 25th Percentile	56	57	59	54	45	58	54	55
Age: Median	65	65	67	66	56	66	62	62
Age: 75th Percentile	72	72	73	73	66	74	70	69
Age: 95th Percentile	82	82	82	83	78	82	80	78
High TMB, >=20 mut/Mb	13%	18%	12%	12%	2%	17%	20%	10%
Intermediate TMB, 6-19 mut/Mb	41%	47%	63%	38%	8%	39%	50%	60%
Low TMB, <=5 mut/Mb	46%	35%	25%	50%	90%	44%	30%	30%

Results

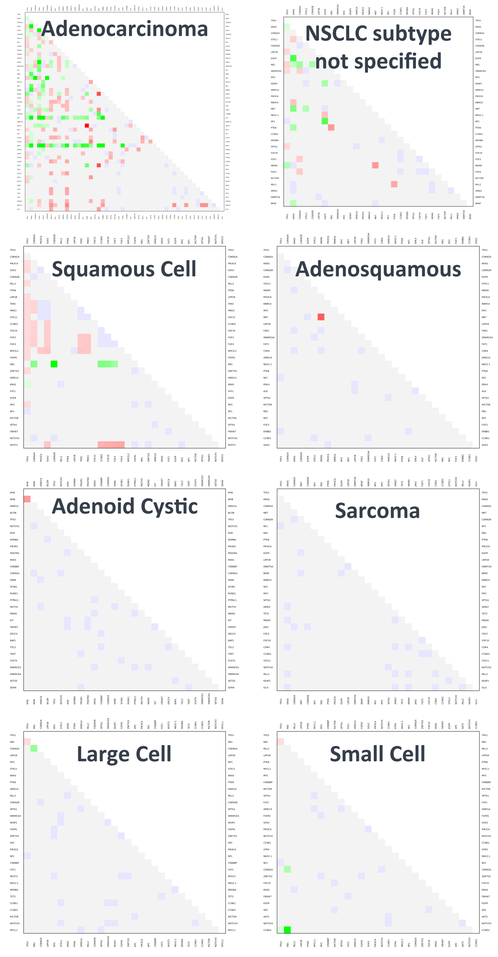
Gene alteration frequencies



Therapeutic actionability



Co-occurrence and mutual exclusivity



Color saturation at 4-fold enrichment or anti-enrichment, p<0.0001

Conclusions

This study presents a comprehensive landscape of the molecular alterations across the most common subtypes of lung cancer, with particular focus on those related to targeted therapy choice. Patients with high TMB are most likely to benefit from immunotherapy, frequently lack other therapeutic targets, and make up a significant fraction of every subtype of lung cancer. The accuracy of comprehensive genomic profiling, coupled with the ability to interrogate all potentially actionable alterations, suggests that this type of testing should be routine component of lung cancer patient care.