zeste homolog 2 (EZH2) histone-lysine methyltransferase in mediating a chemoresistant phenotype through silencing of the SLFN11 gene product, a factor implicated in DNA damage repair deficiency. Furthermore, transient gene expression changes in survival cell fractions following chemotherapy have been demonstrated to contribute to disease relapse and can potentially be targeted. Given the exceptional initial response rates SCLC has to cisplatin and etoposide, we endeavored to define molecular changes that occur in surviving cell fractions following initial chemotherapy challenge to refine our understanding of SCLC relapse biology and identify candidate factors. We initially identified optimal dosing schemes across a panel of SCLC cell lines and quantified cell number and proliferation, establishing seven to ten days as a time window for maximal cytoreduction following chemotherapy in vitro. We then performed transcriptional profiling via RNA-sequencing on cell lines treated with either single-agent cisplatin or combination cisplatin + etoposide across a 24-day time course and utilized principal component analysis to identify genes whose expression exhibits transient expression patterns across the time course. Using gene set enrichment analysis, we confirmed fidelity of our dataset by identification of expected transiently downregulated genes involved in ribosomal biogenesis and concordantly upregulated genes involved in xenobiotic response and DNA damage. Consistently, between both single-agent cisplatin and combination treatment time courses, we identified a significant transient upregulation of a suite of transcription factors. Importantly, we observed a 10- to 30-fold upregulation of these factors compared to baseline that is transient and peaks at timepoints with lowest absolute viable cell number. Current work is focused on determining the sufficiency and necessity of these factors in the progression of SCLC following initial chemotherapy.

B07
Mechanisms of Alectinib Resistance in a Leptomeningeal Carcinomatosis of EML4-ALK Lung Cancer and Its Circumvention by EGR-TKIs

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Central nervous system (CNS) metastasis, such as brain metastasis and leptomeningeal carcinomatosis (LMC), occurs in 20–40% of all patients with cancer. Anaplastic lymphoma kinase (ALK) is a clinically validated drug target, and ALK rearrangements are found in approximately 3–5% of non-small cell lung cancer (NSCLC). ALK tyrosine kinase inhibitor (TKI) shows dramatic clinical efficacy in ALK-rearranged NSCLC patients, and the second-generation ALK-TKI alectinib is effective against CNS metastasis of ALK-rearranged NSCLC. However, the patients with ALK-rearrangement acquire resistance to alectinib over time and develop recurrent LMC metastasis. This study aimed to clarify the mechanism of resistance to alectinib in LMC and seek a novel therapeutic strategy. Alectinib-resistant cell line (A925L/AR) was established by continuous treatment with alectinib in the LMC mouse model inoculated with the alectinib-sensitive human lung cancer cell line, A925LPE3, which harbors the EML4-ALK gene fusion. The tumor level was measured by in vivo imaging system. To clarify the mechanism of alectinib resistance, tumor cell culture supernatants, patient cerebrospinal fluid (CSF), and patient serum were measured using ELISA kits for EGFR ligands. A925L/AR cells were moderately resistant to various ALK-TKIs, such as alectinib, crizotinib, ceritinib, and lorlatinib, compared with parental cells in vitro. A925L/AR cells acquired resistance through epidermal growth factor receptor (EGFR) activation due to overexpression of its ligand, amphiregulin, via inhibited expression of microRNA 449-a. EGFR-TKIs and anti-EGFR antibodies sensitized A925L/AR cells to alectinib in vitro. In the LMC model with A925L/AR cells, combined treatment with alectinib and an EGFR-TKI, such as erlotinib and osimertinib, successfully controlled LMC progression. Imaging mass spectrometry showed accumulation of EGFR-TKIs in the tumor lesions. Moreover, notably high amphiregulin levels were detected in the cerebrospinal fluid from ALK-rearranged NSCLC patients with alectinib-resistant LMC compared with those in EGFR-mutated NSCLC patients with EGFR-TKI-resistant LMC or patients without LMC. We demonstrated that EML4-ALK lung cancer cells acquired moderate resistance to alectinib in the leptomeningeal space due to amphiregulin-triggered EGFR activation. Moreover, combined use of alectinib and EGFR-TKIs, including the third-generation inhibitor osimertinib, could overcome resistance in the LMC model. Our findings may provide rationale for clinical trials to investigate the effects of novel therapies dual-targeting ALK and EGFR in ALK-rearranged NSCLC with alectinib-resistant LMC.

B08
Impact of Concurrent STK11 Loss and c-MYC Amplification in Metastatic Non-Small Cell Lung Cancer (NSCLC)

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Introduction: Despite significant therapeutic advances, clinical outcome remains poor in most patients (pts) with NSCLC, due at least in part to their genotype. STK11 is a master kinase that controls cellular metabolism, while c-MYC is an oncogene altered in many cancers promoting proliferation. Preclinical data (PMID:24793789) suggest that c-MYC amplification in the setting of STK11 loss can lead to unchecked growth of cancer cells. We anecdotally observed rapid progression, primary treatment refractoriness, and dramatic clinical decline in several pts with metastatic NSCLC (mNSCLC) with concurrent STK11 loss and c-MYCamplification. Hence, we investigated the incidence and the prognostic impact of these biomarkers in mNSCLC. Methods: This study was performed through the Precision Medicine Exchange Consortium (PMEC), a consortium of 10 US academic medical centers that share clinically annotated genomic data under a central IRB-approved protocol. The PMEC database (PMEC-DB) was queried for NSCLC pts with either STK11 loss (cohort A), c-MYC amplification (cohort B), or both (cohort C). Comprehensive genomic profiling (CGP) was performed on tumor tissue utilizing the Foundation One 315 gene assay. Demographic and disease characteristics were analyzed. Survival curves were estimated using the Kaplan-Meier method. Results: Among the 1,952 pts with NSCLC in the PMEC-DB, 396 pts met the inclusion criteria with 246 (62%), 103 (26%), and 47 (11.8%) pts in cohorts A, B, and C respectively. Median TMB for the entire study set was 8.7; there was no statistically significant difference between the 3 cohorts (p = 0.12). KRAS mutations were detected more frequently in cohort A compared to cohorts B and C (58% vs. 18% vs 38%; p<0.0001). Clinical outcome data were available in 99 (25%) pts and were distributed among cohorts A, B, and C, in similar proportion to the overall study set with 60, 24, and 15 pts, respectively. Cohort C was associated with a nonadenocarcinoma histology compared to cohorts A and B (53.3%, 16.7%, and 33.3%, respectively, p = 0.011). Nonadenocarcinoma subtypes in Cohort C were NSCLC NOS 33.3%, squamous 6.7%, and large cell neuroendocrine 13.3%. There was no difference in median overall survival (mOS) between cohorts A, B, and C (10 months, 17 months, and 11 months respectively, p =0.68).