mechanism that emerges. Using RNA-seq data, we searched for epigenetic regulators that might be mediating the differentially expressed genes in the resistant cells. This analysis revealed that the chromatin remodeling protein SMARCA4/BRG1 is required for maintenance of the resistant phenotype in one of the models as knockdown of BRG1 sensitized cells to osimertinib. Further analysis revealed that SMARCA4 is stabilized in TKI-resistant cells, thus leading to TKI resistance. Finally, immunohistochemistry (IHC) examination of a collection of TKI-resistant patient-derived xenografts (PDXs) revealed higher levels of SMARCA4 expression in TKI-resistant tumors without on-target EGFR-dependent resistant mechanisms. To further elucidate the role of SMARCA4, we are currently performing ATAC-seq experiments that will offer insights into chromatin accessibility mediated by the protein in the resistant cells. In addition, we are assessing the protein levels of SMARCA4 in clinical specimens obtained before treatment and at the time of resistance by IHC. As new and better targeted therapies are developed, complex resistance mechanisms that involve epigenetic changes in tumors are likely to be increasingly observed. Our studies offer insights into the mechanisms that underlie such resistance that could lead to new therapeutic possibilities for these tumors.

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**Drug Sensitivity and Allele Specificity of First-Line Osimertinib Resistance EGFR Mutations**

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Osimertinib, a mutant-specific third-generation EGFR TKI, is emerging as the preferred first-line therapy for EGFR mutant lung cancer. Despite initial responses in patients, however, resistance inevitably develops over time. In order to investigate mechanisms of resistance to first-line osimertinib, we modeled acquired resistance to this drug in transgenic mouse models of EGFR exon 20-induced lung adenocarcinoma and found that it is mediated largely through secondary mutations in EGFR — either C797S or L718V/Q. Analysis of circulating free DNA data from patients with EGFR mutant lung cancer revealed that L718Q/V mutations almost always arise in the context of an L858R driver mutation, and may occur at least as frequently as C797S in T790M-negative tumors. Therapeutic testing in mice revealed that both erlotinib and afatinib caused repression of osimertinib-resistant C797S-containing tumors, whereas only afatinib was effective in L718Q mutant tumors. Combination first-line osimertinib plus erlotinib treatment prevented the emergence of secondary mutations in EGFR. Finally, we report a patient with a tumor harboring both the L718V and L718Q mutations who benefited from afatinib treatment. Our data identify specific secondary EGFR mutations as a major mechanism of acquired resistance to first-line osimertinib treatment and highlight potential strategies to overcome or prevent osimertinib resistance in vivo. Furthermore, these findings emphasize how knowledge of the specific characteristics of resistance mutations is important for determining potential subsequent treatment approaches.

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**Short-Term Exposure to REV-5901 Decreases the Viability of Chemotherapy-Resistant Adherent Lung Cancer Cells and Floating Tumorspheres**

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Toxicity to normal cells as well as specificity of the presence of highly resistant cancer cells, such as cancer stem-like cells (CS-LCs), are key factors that limit the efficacy of chemotherapy. In tumors, CS-LCs are often associated with chemoresistance and tumor relapse. In this study we used