

## Drug Resistance

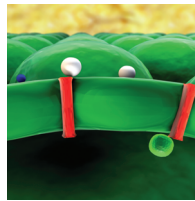
**Major finding:** An allosteric EGFR inhibitor cooperates with cetuximab to selectively target EGFR T790M and C797S.

**Concept:** EAI045 binds an allosteric pocket in T790M-mutant EGFR in a non-ATP-competitive manner.

**Impact:** Allosteric agents may synergize with other/ATP-competitive EGFR inhibitors to limit drug resistance.

### ALLOSTERIC EGFR INHIBITORS OVERCOME RESISTANCE MUTATIONS

Non-small cell lung carcinomas (NSCLC) with activating mutations in the EGFR kinase domain can be treated with tyrosine kinase inhibitors (TKI). Secondary mutations in the ATP site of the receptor, most commonly the T790M mutation, confer resistance by enhancing the affinity for ATP, thereby reducing the potency of ATP-competitive TKIs. EGFR T790M can be targeted by mutant-selective third-generation inhibitors that covalently bind C797 in the ATP binding pocket; however, acquisition of C797 mutations can blunt the efficacy of these agents. As an alternative strategy to target drug-resistant EGFR mutants, Jia and colleagues screened a large compound library to identify allosteric EGFR inhibitors that could potentially overcome the increased ATP affinity of EGFR T790M. EGFR allosteric inhibitor-1 (EAI001) was identified as a selective, non-ATP-competitive inhibitor of EGFR T790M and L858R. Crystal structure analysis revealed that EAI001 bound to an allosteric pocket of EGFR T790M formed by displacement of the C-helix in the inactive kinase conformation. EAI001 bound to the mutant EGFR gatekeeper methionine residue, but did not bind the inactive wild-type EGFR conformation, conferring specificity for mutant EGFR. Compound optimization generated EAI045, an



L858R/T790M inhibitor with enhanced potency and selectivity against mutant EGFR. However, EAI045 only partially inhibited EGFR phosphorylation and cell proliferation in L858R/T790M-mutant NSCLC cells; EGF-induced EGFR dimerization rendered the allosteric binding pocket differentially accessible in the two subunits of the asymmetric dimer, limiting the efficacy of single-agent EAI045. In contrast, blocking dimerization with the therapeutic anti-EGFR antibody cetuximab enhanced EAI045 potency. In a genetically engineered mouse model of mutant EGFR L858R/T790M-driven lung cancer, EAI045 synergized with cetuximab to promote tumor regression. Further, the combination was effective against L858R/T790M/C797S-mutant tumors that are resistant to third-generation EGFR inhibitors. Taken together, these results demonstrate that targeting allosteric sites can generate mutation-specific TKIs, and suggest that combined treatment with EAI045 and cetuximab or ATP-competitive EGFR inhibitors may warrant further clinical investigation in NSCLC. ■

Jia Y, Yun C-H, Park E, Ercan D, Manuia M, Juarez J, et al. Overcoming EGFR(T790M) and EGFR(C797S) resistance with mutant-selective allosteric inhibitors. *Nature* 2016;534:129–32.

## Hepatocellular Carcinoma

**Major finding:** TP53-altered HCC tumors become addicted to MYC stabilization.

**Mechanism:** AURKA binds to and stabilizes phospho-MYC, overcoming G2/M cell-cycle arrest in TP53-altered cells.

**Impact:** AURKA conformation-changing inhibitors may be beneficial in patients with TP53-altered HCC.

### DISRUPTING THE MYC-AURKA INTERACTION MAY SUPPRESS p53-ALTERED TUMORS

MYC is upregulated in many human tumors and is implicated in tumor initiation and maintenance, but is considered an undruggable target. Thus, strategies to target MYC indirectly are of interest. Proteolytic turnover of the closely related MYCN protein has been shown to be regulated by a kinase-independent function of aurora kinase A (AURKA), and inhibitors that disrupt the native conformation of AURKA can induce MYCN degradation. However, it is not clear if MYC-AURKA complexes exist and could be therapeutically targeted. An *in vivo* shRNA screen in a mouse model of *Nras*<sup>G12V</sup>; *Trp53*<sup>-/-</sup> hepatocellular carcinoma (HCC) performed by Dauch and colleagues revealed that *Aurka* knockdown can override p19<sup>ARF</sup>-dependent G2/M cell-cycle arrest in hepatocytes, increase tumor development, and reduce overall survival, indicating a tumor-suppressive role of AURKA in this context. However, in the context of chronic liver damage, both AURKA and MYC expression were high in *Trp53*<sup>-/-</sup> tumors, and MYC regulated cell-cycle reentry in *Trp53*<sup>-/-</sup> hepatocytes. MYC phosphorylation is required for its degradation, and AURKA bound to and stabilized phospho-MYC, overcoming G2/M cell-cycle arrest and promoting tumor cell survival.

Alisertib, an AURKA inhibitor in clinical development that induces a conformational change in AURKA, resulted in MYC degradation and increased survival in HCC MYC-overexpressing *Nras*<sup>G12V</sup>; *Trp53*<sup>-/-</sup> mice, whereas AURKA inhibitors that did not induce a conformational change did not have an effect on *Trp53*-altered cells. Further, in a human HCC cohort, mutant *TP53* was associated with increased mRNA expression of AURKA and of MYC target genes, and treatment of xenografted human *TP53*-altered HCC cell lines with alisertib resulted in tumor regression, whereas the growth of *TP53* wild-type HCC cell line xenografts was not significantly affected. Altogether, these findings suggest that conformation-changing AURKA inhibitors may be used to indirectly target MYC, and indicate that further clinical investigation of AURKA conformation inhibitors may be warranted for the treatment of *TP53*-altered tumors. ■

Dauch D, Rudalska R, Cossa G, Nault J-C, Kang T-W, Wuestefeld T, et al. A MYC-aurora kinase A protein complex represents an actionable drug target in p53-altered liver cancer. *Nat Med* 2016 May 23 [Epub ahead of print].