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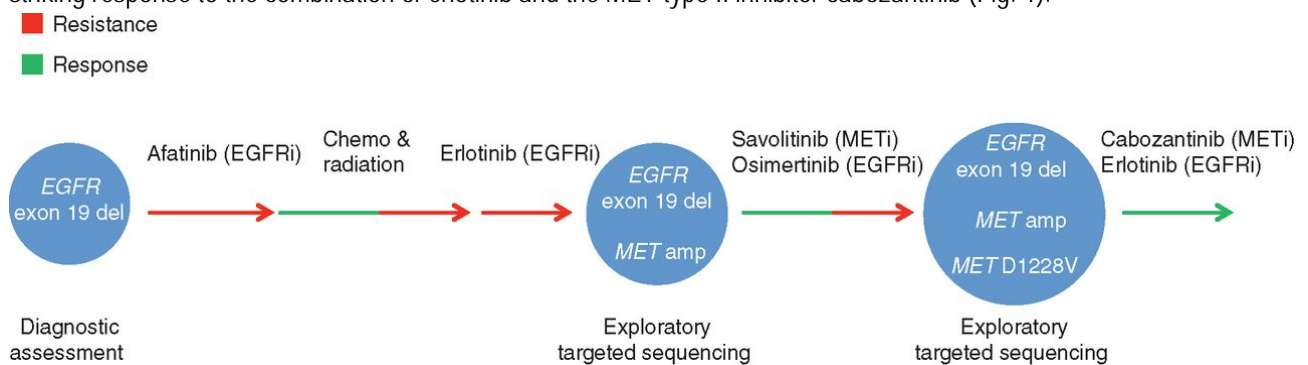
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# Oncogenic MET as an Effective Therapeutic Target in Non-Small Cell Lung Cancer Resistant to EGFR Inhibitors: The Rise of the Phoenix

Livio Trusolino

Non-small cell lung cancers (NSCLC) harboring EGFR-activating mutations, such as exon 19 deletion, account for approximately 10% to 12% of all NSCLCs in Caucasians and usually respond better to anti-EGFR therapies than tumors with wild-type EGFR. However, in up to 20% of cases, the selective pressure exerted by EGFR blockade leads to the emergence of cellular subclones harboring amplification of the MET oncogene, encoding the tyrosine kinase receptor for hepatocyte growth factor. This increase in MET gene copy number results in kinase overexpression and constitutive activation, with the ensuing propagation of downstream survival signals that substitute for EGFR and impose resistance to EGFR inactivation. The MET-amplified subclones may either preexist in the original tumor population because of genetic heterogeneity or be induced de novo by treatment in a stochastic fashion, due to inherent genomic instability (1, 2). Preclinical studies in cell lines and animal models have demonstrated that pharmacologic interception of MET in this specific genetic context reverts resistance and restores sensitivity to EGFR inhibition (1, 2).

In this issue of *Cancer Discovery*, Bahcall and colleagues (3) describe a patient with metastatic NSCLC positive for an EGFR exon 19 deletion mutation, who did not respond to EGFR neutralization by first-generation (erlotinib) or second-generation (afatinib) inhibitors. Targeted massively parallel sequencing of a biopsy from a growing cervical lymph node identified high-grade amplification of MET, in addition to the known EGFR exon 19 deletion. Similar to other tyrosine kinase inhibitors, small molecules targeting MET fall into two functionally distinct categories: Type I inhibitors typically interact with the ATP-binding site of the active form of the kinase; conversely, type II inhibitors display only partial interaction with the ATP-binding cleft and extend into an adjacent allosteric pocket that is exposed exclusively by the inactive kinase conformation. When given combination therapy with a type I MET inhibitor (savolitinib) and a mutant-selective EGFR inhibitor (osimertinib), the patient experienced an initially dramatic clinical response, but eventually progressed with a new nodule in the lung. A second sequencing analysis on the recurrent lung metastasis revealed an acquired D1228V (3683A>T) variant in the MET kinase domain, present at a high allelic fraction (43%). Based on protein structural studies, the authors elucidate MET<sup>D1228V</sup>-mediated resistance to savolitinib (and other type I MET inhibitors) as a mutation-induced repositioning of the kinase activation loop, which however retains binding to type II inhibitors. In biochemical assays, type II, but not type I, MET inhibitors were able to suppress kinase phosphorylation and downstream signaling in NSCLC cells ectopically expressing the D1228V mutation. Consistently, the patient had a striking response to the combination of erlotinib and the MET type II inhibitor cabozantinib (Fig. 1).



**Figure 1.**

Molecular and clinical history of case patient, as reported by Bahcall and colleagues (3). The timeline is not scaled. EGFRi, EGFR inhibitor; METi, MET inhibitor; del, deletion; amp, amplification.

The first piece of information presented by Bahcall and colleagues (3) is that a combination of therapies against EGFR and MET has anticancer activity in NSCLCs concomitantly carrying an EGFR mutation and a MET amplification. These data add on a recent case study showing, again, substantial tumor regression after EGFR and MET coinhibition in another patient with NSCLC with a lesion sharing the same genetic makeup (4). Although anecdotal, the two reports

bring to the fore MET as a viable therapeutic target and relaunch the deployment of MET inhibitors in lung cancer. This is much welcome—and much needed—news after the negative phase III trials with onartuzumab, an anti-MET monoclonal antibody that prevents ligand binding, and tivantinib, a putative MET small-molecule inhibitor that also exerts MET-independent microtubule-disrupting activity. The two studies were large randomized efforts evaluating onartuzumab (The METLung trial; ref. 5) or tivantinib (MARQUEE; ref. 6) in combination with erlotinib in previously treated patients with advanced NSCLC, and both were discontinued early for lack of clinically meaningful activity after a prespecified interim analysis. In hindsight, and when considering the proved efficacy of EGFR–MET cotargeting in patients with EGFR-mutant, MET-amplified NSCLC, the reasons for these failures can be easily explained: The only criterion for patient inclusion in both trials was MET-positive expression by immunohistochemistry, and no up-front evaluation of EGFR and MET genetic status was planned. Let alone the hurdles and challenges of defining dichotomous thresholds for continuous variables with a normal distribution—like semi-quantitative assessment of protein expression in tissues—a transmissible and selectable genetic change such as MET amplification is likely to constitute a stronger oncogenic event than MET overexpression only, and much more poised to sustain tumor reliance on MET signaling (7).

Learning from these lessons, it is now evident that future clinical studies in the field need to be rethought using an obligate selection of patients with EGFR-mutant tumors and assessment of MET gene amplification as procedural imperatives. Rationally based trials should entail randomization of patients with EGFR-mutant NSCLC for treatment with anti-EGFR and anti-MET compounds to investigate whether blockade of MET *ab initio* can contrast outcompetition by incipient MET-amplified subpopulations, hence delaying the onset of resistance. Other sensible approaches should also be contemplated that systematically analyze MET gene amplification (not MET protein expression) in patients with EGFR-mutant NSCLC who relapse on erlotinib or other EGFR inhibitors; in this way, MET-amplified refractory cases could be reallocated to dual treatment with EGFR- and MET-targeting agents to explore whether MET inhibition, and subsequent resensitization to EGFR blockade, can regress tumors with an acquired preponderance of MET-amplified clones. It is likely that onartuzumab will not be the optimal anti-MET agent to contrast the growth of MET-amplified tumors, because it mainly acts by displacing ligand-receptor binding and not by impairing MET catalytic activity; likewise, the specificity of tivantinib in inhibiting MET is a matter of debate, which raises a word of caution on data interpretation as a whole. This said, the premature termination of METLung and MARQUEE should not be taken as a warning against the value of MET as a key target in EGFR-mutant lung adenocarcinoma, but rather as the consequence of imperfect choice of response biomarkers for patient selection. It is also fair to say that using genetic profiling to enrich for potential responders may be logistically laborious, given the low frequency of EGFR and MET aberrations in NSCLC. The implementation of umbrella trials, whereby a common genomic screening platform is utilized to detect multiple DNA alterations of predicted significance in a given tumor type, is expected to facilitate swift identification of patients with tumors bearing rare genetic anomalies.

The patient described in ref. 3 responded successfully to the anti-EGFR/anti-MET combination therapy, but after 8 months developed resistance. The second piece of information provided by Bahcall and colleagues (3) is the identification of the genetic cause of this resistance: The treatment-refractory metastasis exhibited a D1228V mutation in the MET gene, which leads to a repositioning of the kinase activation loop and thus prevents binding of type I inhibitors. This alteration and the description of its functional consequences are not new: Mutations at the D1228 residue were initially detected in cancer cell lines upon a mutagenesis-based resistance screen, and found to cause desensitization to type I but not type II MET inhibitors (8). Intriguingly, the same mutation has been recently identified in a patient with NSCLC displaying a MET-activating exon skipping variant and acquired resistance to the type I MET inhibitor crizotinib (9). Although this observation highlights the pervasive nature of such resistance-conferring mutations in MET-driven NSCLCs, irrespective of the original MET abnormality, it remains descriptive in the absence of a successful strategy to tackle the mutant protein therapeutically; the merit of the study by Bahcall and colleagues (3) is indeed illustrated by the striking response experienced by the case patient to the combination of erlotinib and the type II MET inhibitor. This outcome motivates the prospective routine testing for D1228 mutations in patients with MET-dependent tumors who are initially sensitive to type I MET inhibitors and then relapse, as such patients could be reassigned to a potentially beneficial treatment with type II MET inhibitors. Of note, the authors (3) succeeded in developing a droplet digital PCR assay for detection of MET<sup>D1228V</sup> in plasma, which supports the feasibility of noninvasive serial genotyping in future accomplishments.

The rational application of MET inhibitors in the clinic, especially in the case of NSCLC, brings with it a history of hopes, falls, and rises. Positive—albeit sporadic—hints are progressively accumulating, including preliminary evidence of sensitivity to anti-MET monotherapy in patients with NSCLC with high-grade MET amplification in the absence of concurrent EGFR mutations or MET exon 14 skipping variants (splicing aberrations that result in the deletion of a negative regulatory domain of the MET kinase; ref. 10). The time is ripe now to move from exceptions to rules: Promising results in individual responders (such as those reported in ref. 3, and the other examples mentioned) should be consolidated on a larger scale; genomic biomarkers of response (MET amplification, exon 14 skipping) and resistance (the MET<sup>D1228V</sup> mutation, and others that will surely surface in the near future) should be put in the context of the coexisting genetic and functional traits that may further improve sensitivity or, conversely, exacerbate therapeutic refractoriness. Ultimately, population-level stratification and high-density molecular annotation of patient responses will enable the development of decisional algorithms that collectively capture the genetic underpinnings of NSCLC susceptibility to MET inhibition as a means to inform therapeutic decisions and dynamically adapt them over the course of treatment.

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No potential conflicts of interest were disclosed.

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