Translational research

1580P THERAPEUTIC TARGETING OF RAF-INDUCED PARADOXICAL ERK ACTIVATION WITH A VERTICAL COMBINATION HITTING MULTIPLE STEPS ALONG THE MAPK CASCADE

U. Cesta Incani, A. Del Curatolo, I. Falcone, A. Eramo, S. Mattone, S. Shirasawa, M. Brogini, R. De Maria, F. Cognetti, L. Ciuffreda, M. Mieli
1Medical Oncology A, Regina Elena National Cancer Institute, Rome, ITALY
2Department of Hematology, Oncology and Molecular Medicine, Istituto Superiore di Sanita, Rome, ITALY
3Biostatistics, Regina Elena National Cancer Institute, Rome, ITALY
4Experimental Oncology, Regina Elena National Cancer Institute, Rome, ITALY
5Department of Cell Biology, Fukuoka University, Fukuoka, JAPAN
6Oncology, Istituto Mario Negri, Milan, ITALY
7 Regina Elena National Cancer Institute, Rome, ITALY

Aim: Mounting evidence suggests that RAF-mediated MEK activation plays a crucial role in paradox MAPK (re)activation, leading to resistance and therapeutic failure with agents hitting a single step along the MAPK cascade.

Methods: We examined the molecular and functional effects of single and combined MEK (trametinib, T), BRAF (dabrafenib, D), and pan-RAF (RAF265, R) inhibition, using WB and conservative isobologram analysis to assess functional synergism and explored genetic determinants of synergistic interactions.

Results: In BRAF-mut models, both D and T effectively inactivated MAPK and inhibited cell growth, with no synergistic growth inhibition with their combination (CI: 0.7-1.3x10^6). Conversely, in KRAS-mut lung (2/5 cell lines) and pancreatic (4/6 cell lines) cancer models, in which selective BRAF inhibition induced paradox hyperphosphorylation of MEK, ERK, and p90RSK, combined D+T synergistically suppressed malignant growth (CI: 0.1-0.7). At concentrations inhibiting all RAF isoforms, R did not induce paradox MAPK activation and did not result in growth inhibitory synergism when combined with T. Highly synergistic in vitro growth inhibition was also observed with D+T in 2/5 patient-derived lung cancer stem cells. KRAS mutations appeared to be necessary, but not sufficient, to determine paradox MAPK activation and functional synergism with D+T, as assessed in two KRAS isogenic cell lines (H1299 expressing individual codon 12 mutants and HCT116 clones differing for homo or heterozygous G13D). Conversely, in lung cancer models driven by EGFR family activation (EGFR-mut HCC827 and H1650 cells and HER-2 overexpressing Calu-3 cells), paradox MAPK activation upon selective BRAF inhibition was dependent on upstream receptor activity and could be efficiently blocked by the EGFR/HER-2 inhibitor Lapatinib.

Conclusions: Overall, our data indicate that RAF-mediated, paradox MAPK activation may be sustained by both upstream (RTK activation) and downstream (KRAS mutations) mechanisms and that in appropriate cellular contexts vertical RAF/MEK inhibition-based combination strategies may exert highly synergistic antitumor effects.

Disclosure: All authors have declared no conflicts of interest.

© European Society for Medical Oncology 2014. Published by Oxford University Press on behalf of the European Society for Medical Oncology. All rights reserved. For permissions, please email: journals.permissions@oup.com.