Editorial

Predicting responsiveness to sorafenib: can the determination of FGF3/FGF4 amplifications enrich for clinical benefit?

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Abstract: A small proportion of patients with advanced hepatocellular carcinoma harbor FGF3/4 gene amplifications in an 11q13 amplicon. A recent report suggests that the presence of this alteration might enrich for sensitivity to sorafenib. Further, there is growing evidence that interference with the FGF/FGFR axis has therapeutic potential in patients with advanced hepatocellular carcinoma.

Keywords: Hepatocellular carcinoma (HCC); fibroblast growth factor (FGF); sorafenib; brivanib; targeted therapy

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In the April 2013 issue of the journal, Arao and colleagues used comparative genomic hybridization (CGH) to interrogate a resected hepatocellular carcinoma (HCC) after a near complete response to sorafenib, 5-fluororacil, and interferon for 14 months (1). Confirming previous findings in HCC, gains of 1q, 8q and a high level amplicon at 11q13 were identified by this analysis (2-4). Given the striking tumor shrinkage in the clinical outlier, the authors proposed that a gene (i.e., *CCND1*, *FGF19*, *FGF3* and *FGF4*) in this amplicon might be responsible for the unusual responsiveness to treatment. Subsequently, retrospective analysis of 48 banked samples (10 responders and 38 nonresponders) showed that 3 of 10 (30%) patients with complete or partial responses by RECIST version 1.0 had amplifications of fibroblast growth factor 3 and 4 (*FGF3/4*) by multiple methods, and that the frequency of this aberration occurred in 2.4% of all HCC samples. *FGF3/4* amplifications were not observed in patients with stable or progressing disease. In two preclinical models, the authors demonstrated that sorafenib led to greater antitumor activity in cell lines harboring *FGF* or fibroblast growth factor receptor (*FGFR*) amplifications as well as in a xenograft murine model derived from *FGF4* overexpressing HCC cell lines. The authors concluded that *FGF3/4* amplifications might be a useful biomarker for sorafenib responsiveness after further prospective validation.

Sorafenib remains the only systemic therapy proven to modestly improve overall survival over best supportive care in patients with advanced HCC (5,6). Objective responses rarely occur (2% of patients) and sorafenib treatment only results in an 11% absolute benefit in disease control over best supportive care. Thus, it is germane to define biomarkers of responsiveness to aid in patient selection. Such predictive biomarkers are elusive; however, due to the non-specific mechanism of drug action, the rarity of objective tumor response in HCC, and the paucity of clinically-annotated pretreatment tumor specimens for correlative analysis (7). The report of Arao and other recent publications are critical milestones for patient care, and perhaps, represent the beginning of precision medicine, a means of using *a priori* tumoral molecular variants to enrich for sensitive HCCs and select out resistant ones (8). Circulating plasma factors and clinicopathologic characteristics have not been fruitful as predictive tools for sorafenib activity (9-11). In contrast, molecular and proteomic characterization of HCCs suggest that enhanced ERK signaling (12) or the presence of a *VEGFA* amplicon (13) are associated with clinical benefit, while mTOR activation (14) leads to relative resistance to sorafenib. In the hypothesis-generating work of Aroa and colleagues, *FGF3/FGF4* amplifications must be considered

as a potential determinant of sorafenib activity.

Although the authors should be commended for an important translational undertaking in HCC biology, several caveats must be acknowledged in their report. First and foremost, the small sample size and retrospective design leads to selection bias, thus as the authors note, prospective validation is required for this novel genomic marker. Second, retrospective determination of tumor shrinkage is difficult due to non-uniformity of technique, timing of scans, and investigator bias (15). Third, the 11q13 amplicon contains a number of other candidate genes and is often associated with other high level amplicons (i.e., 8p), whose gene products might be affected by sorafenib and were not assayed for in these experiments (16). That is, although it is rationale to assay for *FGF3/4* copy number after its identification in an impressive index case, the observation that *FGF3/4* is amplified may merely be an association and not mechanistically causal for sorafenib sensitivity. Other investigations indicate that although *FGF3/4* is amplified in HCC, its presence does not correlate with a reciprocal increase in gene expression (3). This suggests that an increase in *FGF3/4* copy number may have little functional significance. It is also important to recognize that the index case was treated with multimodality therapy, thus it is possible that the observed response, as noted by the authors, might represent a mixed effect. Fourth, the authors do not acknowledge that tumor shrinkage is observed (albeit rarely) in patients undergoing best supportive care. Is it possible that the *FGF3/4* amplification might represent a prognostic marker, portending more favorable disease biology? Available evidence would suggest that this is not the case as alterations in FGF pathways have been associated with more aggressive clinical parameters (17,18). Finally, the authors do not reconcile the observation that increased FGF signaling has been hypothesized as a mechanism of anti-angiogenic escape to sorafenib, with preclinical data indicating that hypoxia induces upregulation of a several members of the FGF family (19,20).

The role of FGF/FGFR blockade as a treatment in HCC is under active investigation and the finding that 2.4% of the study population harbor alterations in *FGF3/FGF4* is of critical importance. Although a small proportion, when accounting for the global disease burden of HCC, this represents a substantial number of patients who might benefit from FGF pathway interference. It is also plausible that the frequency of this alteration may vary based on HCC etiologic agent or as suggested by the authors, might be enriched in patients with the clinical phenotype of pulmonary metastasis. In addition, other pathway aberrations, such as activating missense mutations or splice variants of FGFR, might be discovered in HCC. Preclinical data indicate that antibodies to FGF-19, a ligand for FGFR4, are an effective therapeutic strategy in cell lines harboring 11q13 amplicons (3) and brivanib, a dual inhibitor of VEGFR2 and FGFR, suppresses HCC tumor growth in xenograft models with a more pronounced effect in FGFR1/2 expressing tumors (21).

Clinical data with brivanib has been humbling but insightful. In a molecularly non-selected advanced HCC patient population, brivanib was not superior or non-inferior to sorafenib in a large, double blind, placebo controlled phase III study in the first line (22). A large, randomized phase III study of brivanib compared to best supportive care in advanced HCC patients who progressed on sorafenib was also negative (23). The primary endpoint of overall survival was not met [median OS 9.4 for brivanib *vs*. 8.2 for placebo, HR =0.89, (95% CI, 0.69-1.15)] though there was an observed benefit in secondary outcomes, such as objective response (10% *vs*. 2%) and time to progression (4.3 *vs*. 2.7). A potential reason for the failure of this study was an underestimation of the effects of best supportive care in the second line setting. Taken together, these data suggest that inhibition of FGFR might have some utility after sorafenib failure but also indicate that specific analysis of tumoral FGF/FGFR pathway aberrations is warranted to select for therapeutic benefit. Data emerging from other compounds known to interfere with FGFR signaling, with the multi-targeted tyrosine kinase inhibitors dovitinib (24) and orantinib (TSU-68) (25), and selective FGFR inhibitors such as Debio1347 (clinicaltrial.gov NCT01948297) and BGJ389 (clinicaltrial.gov NCT02160041) will be insightful and may represent a new avenue for therapy in HCC.

In summary, the report of Arao and colleagues, is intriguing and is one of the first instances of an alteration in the HCC genome that suggests benefit to a therapeutic agent in HCC. At Memorial Sloan Kettering, advanced HCC patients with available peripheral blood and tumoral tissue may participate in a prospective genotyping effort. Paired samples undergo solution phase hybridization-based exon capture and massively parallel DNA sequencing to capture all protein-coding exons and select introns in over 340 oncogenes, tumor suppressor genes, and members of pathways deemed actionable by targeted therapies (26,27). This assay can identify three classes of somatic alterations: single-nucleotide variants, small insertions/deletions

(indels), and copy number alterations to help inform HCC clinical trial participation in the era of targeted therapeutics. Although more investigation is required, the identification of *FGF3/4* amplifications is another step in our path to improve therapies in this disease and a potential means to select patients for clinical benefit.

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References

1. Arao T, Ueshima K, Matsumoto K, et al. FGF3/FGF4 amplification and multiple lung metastases in responders to sorafenib in hepatocellular carcinoma. Hepatology 2013;57:1407-15.
2. Moinzadeh P, Breuhahn K, Stützer H, et al. Chromosome alterations in human hepatocellular carcinomas correlate with aetiology and histological grade-results of an explorative CGH meta-analysis. Br J Cancer 2005;92:935-41.
3. Sawey ET, Chanrion M, Cai C, et al. Identification of a therapeutic strategy targeting amplified FGF19 in liver cancer by Oncogenomic screening. Cancer Cell 2011;19:347-58.
4. Chiang DY, Villanueva A, Hoshida Y, et al. Focal gains of VEGFA and molecular classification of hepatocellular carcinoma. Cancer Res 2008;68:6779-88.
5. Llovet JM, Ricci S, Mazzaferro V, et al. Sorafenib in advanced hepatocellular carcinoma. N Engl J Med 2008;359:378-90.
6. Cheng AL, Kang YK, Chen Z, et al. Efficacy and safety of sorafenib in patients in the Asia-Pacific region with advanced hepatocellular carcinoma: a phase III randomised, double-blind, placebo-controlled trial. Lancet Oncol 2009;10:25-34.
7. Harding JJ, Abou-Alfa GK. Treating advanced hepatocellular carcinoma: how to get out of first gear. Cancer 2014. [Epub ahead of print].
8. Garraway LA, Verweij J, Ballman KV. Precision oncology: an overview. J Clin Oncol 2013;31:1803-5.
9. Llovet JM, Peña CEA, Lathia CD, et al. Plasma biomarkers as predictors of outcome in patients with advanced hepatocellular carcinoma. Clin Cancer Res 2012;18:2290-300.
10. Cheng AL, Guan Z, Chen Z, et al. Efficacy and safety of sorafenib in patients with advanced hepatocellular carcinoma according to baseline status: Subset analyses of the phase III Sorafenib Asia-pacific trial. Eur J Cancer 2012;48:1452-65.
11. Bruix J, Raoul JL, Sherman M, et al. Efficacy and safety of sorafenib in patients with advanced hepatocellular carcinoma: subanalyses of a phase III trial. J Hepatol 2012;57:821-29.
12. Abou-Alfa GK, Schwartz L, Ricci S, et al. Phase II study of sorafenib in patients with advanced hepatocellular carcinoma. J Clin Oncol 2006;24:4293-300.
13. Horwitz E, Stein I, Andreozzi M, et al. Human and mouse VEGFA-amplified hepatocellular carcinomas are highly sensitive to sorafenib treatment. Cancer Discov 2014;4:730-43.
14. Masuda M, Chen WY, Miyanaga A, et al. Alternative mammalian target of rapamycin (mTOR) signal activation in sorafenib-resistant hepatocellular carcinoma cells revealed by array-based pathway profiling. Mol Cell Proteomics 2014;13:1429-38.
15. Sharma MR, Maitland ML, Ratain MJ. RECIST: no longer the sharpest tool in the oncology clinical trials toolbox—point. Cancer Res 2012;72:5145-49.
16. Wilkerson PM, Reis-Filho J. The 11q13-q14 amplicon: clinicopathological correlations and potential drivers. Genes Chromosomes Cancer 2013;52:333-55.
17. Mise M, Arii S, Higashituji H, et al. Clinical significance of vascular endothelial growth factor and basic fibroblast growth factor gene expression in liver tumor. Hepatology 1996;23:455-64.
18. Lai JP, Sandhu DS, Yu C, et al. Sulfatase 2 up-regulates glypican 3, promotes fibroblast growth factor signaling, and decreases survival in hepatocellular carcinoma. Hepatology 2008;47:1211-22.
19. Bergers G, Hanahan D. Modes of resistance to anti-angiogenic therapy. Nat Rev Cancer 2008;8:592-603.
20. Casanovas O, Hicklin DJ, Bergers G, et al. Drug resistance by evasion of antiangiogenic targeting of VEGF signaling in late-stage pancreatic islet tumors. Cancer Cell 2005;8:299-309.
21. Huynh H, Ngo VC, Fargnoli J, et al. Brivanib alaninate, a dual inhibitor of vascular endothelial growth factor receptor and fibroblast growth factor receptor tyrosine kinases, induces growth inhibition in mouse models of human hepatocellular carcinoma. Clin Cancer Res 2008;14:6146-53.
22. Johnson PJ, Qin S, Park JW, et al. Brivanib versus sorafenib as first-line therapy in patients with unresectable,

advanced hepatocellular carcinoma: results from the randomized phase III BRISK-FL study. J Clin Oncol 2013;31:3517-24.
23. Llovet JM, Decaens T, Raoul JL, et al. Brivanib in patients with advanced hepatocellular carcinoma who were intolerant to sorafenib or for whom sorafenib failed: results from the randomized phase III BRISK-PS study. J Clin Oncol 2013;31:3509-16.
24. Chen ZY, Shi M, Peng LX, et al. Dovitinib preferentially targets endothelial cells rather than cancer cells for the inhibition of hepatocellular carcinoma growth and metastasis. J Transl Med 2012;10:245.
25. Kanai F, Yoshida H, Tateishi R, et al. A phase I/II trial of the oral antiangiogenic agent TSU-68 in patients with advanced hepatocellular carcinoma. Cancer chemother pharmacol 2011;67:315-24.
26. Catalanotti F, Solit DB, Pulitzer MP, et al. Phase II trial of MEK inhibitor selumetinib (AZD6244, ARRY-142886) in patients with BRAFV600E/K-mutated melanoma. Clin Cancer Res 2013;19:2257-64.
27. Wagle N, Berger MF, Davis MJ, et al. High-throughput detection of actionable genomic alterations in clinical tumor samples by targeted, massively parallel sequencing. Cancer Discov 2012;2:82-93.

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