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Pathways of Hepatocellular Carcinoma Kinase in Epithelial-Mesenchyme Transition

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Introduction

Hepatocellular Carcinoma (HCC) is a difficult-to-treat malignancy that has no treatable genetic mutations. Systemic treatments for advanced HCC are ineffectual, necessitating the identification of prognostic biomarkers and pharmacological targets. The majority of HCC drugs target protein kinases, demonstrating that kinase dependent signalling networks are at the root of HCC formation. We employed a pharmacoproteomics method to link kinome activity in 17 HCC cell lines with their responses to 299 Kinase Inhibitors (KIs) to uncover HCC signalling networks that determine KI responses, yielding a comprehensive set of pathway-based drug response signatures.. By analysing patient HCC data, we established clinical HCC treatment response patterns in individual tumours. Our findings reveal kinase networks that promote EMT and drug resistance, including a FZD2-AXL-NUAK1/2 signalling module [1].

Description

Hepatocellular Carcinoma (HCC) is the world's fourth highest cause of cancer-related death (Global Burden of Disease Cancer Collaboration 2017), and it can be caused by a number of things, including viral hepatitis, alcoholic cirrhosis, and Nonalcoholic Steatohepatitis (NASH) Because HCC exhibits one of the fewest drug gable genetic alterations among solid tumours, treatment options for advanced HCC are limited. The Cancer Genome Atlas Research Network, 2017Five of the seven FDA approved therapies for advanced HCC, including the small molecule medicines Sorafenib Regorafenib Lenvatinib and Cabozantinib and the antibody Ramucizumab (Abou Predictive biomarkers, on the other hand, are a type of biomarker that may be used to predict the outcome of aIn HCCs that initially react to therapy, drug resistance develops; this has been extensively reported for Sorafenib, and shows that HCCs engage compensatory signalling pathways to drive rebound development). The Epithelial-Mesenchymal Transition (EMT), which is the interconversion of an epithelial-like cancer cell phenotype to a mesenchymal-like cancer cell phenotype, is linked to several of these compensatory mechanisms in carcinomas under physiological conditions, the EMT is a crucial part of tissue development and repair). Cancer, on the other hand, hijacks cell signalling networks that control the EMT to enhance tumour progression and drug resistance. Hepatocellular Carcinoma (HCC) is the world's fourth leading cause of cancer-related mortality. Protein kinases are crucial components of most cell signalling networks, and their function is commonly disturbed in cancer. Despite the fact that particular kinases have been shown to have a role in phenotypic transition (Gay there has been no comprehensive mapping of EMT-associated kinase pathways. We investigated HCC responses to kinase inhibitor medications using a kinome-centric pharmacoproteomics strategy, reasoning that studying unknown and dysregulated HCC kinase signaling could reveal new mechanisms of EMT-related drug resistance, molecular indicators of drug response, and new therapeutic targets [2].

In HCCs that respond to treatment initially, drug resistance develops; this has been commonly observed for Sorafenib. Researchers must analyse kinase expression levels, Post-Translational Modifications (PTMs), and their linkages with regulatory proteins to determine the functioning of kinase-dependent signaling networks. All MS-based proteomics techniques can be used to assess kinase expression and function, including global proteomics and phosphoproteomics targeted MS analyses of kinase activation loop phosphorylation sites and kinobead kinase affinity enrichment coupled to phosphopeptide analyses (kinobead/LC-MS)). Kinobead/LC-MS covers kinases, their PTMs, and kinase-regulatory proteins comprehensively and unbiasedly, allowing for kinome activity monitoring). It's necessary to quantify kinase expression levels, Post-Translational Modifications (PTMs), and their relationship to regulatory proteins. Hepatocellular Carcinoma (HCC) is the fourth most common cancer-related cause of death worldwide. In most cell signaling networks, protein kinases are the core nodes.. We used kinobead/LC-MS to map characteristics of kinome activity to growth suppression caused by 299 kinase-targeted medications across a panel of 17 HCC cell lines. We assessed the connection of 275 kinase-dependent cancer pathways with treatment responses using Gene Set Enrichment Analysis (GSEA), resulting in a comprehensive database of KI medication response signatures in HCC We employed kinobead/LC-MS to evaluate drug response signatures in human HCC samples, which might be used as prognostic markers for individualised therapy. Quanti Hepatocellular Carcinoma (HCC) is the fourth most common cancer related cause of death worldwide. Our findings demonstrated that the cellular EMT state had a broad impact on kinase expression and activation, as well as other signaling networks and drug response characteristics, in the majority of instances. In HCC cells, we uncovered a FZD2-AXL-NUAK1/2 signaling pathway that causes EMT. After genetic knockdown or small molecule inhibition of these proteins, the EMT was reversed, replication stress signaling was amplified, and HCC cells were more susceptible to treatments. Hepatocellular Carcinoma (HCC) is the fourth most common cause of cancer related death worldwide. In most cell signaling networks, protein kinases are the core nodes. We used kinobead/LC-MS to map characteristics of kinome activity to growth suppression caused by 299 kinase-targeted medications across a panel of 17 HCC cell lines. To determine the strength of a gene's HCC Pharmacoproteomics Study with a Kinome-Centric Approach relationship Results [3].

We used our kinobead/LC-MS technique to evaluate kinase expression, phosphorylation states, and kinase-interacting proteins in a panel of 17 HCC lines from the Cancer Cell Line Encyclopedia (CCLE) with varied kinase mRNA expression. Using functionally defined phosphosites, kinase-interacting proteins, and their phosphorylation sites, the activation states of 284 of the 346 kinases were identified (see STAR Methods). We used a 299-member diversity library of experimental, pre-clinical, and clinical KIs, all of which inhibit at least one other KI. We observed different responses to inhibitors using the area under the dose-response curve (AUC) as a measure of pharmacological effectiveness. FGFR, EGFR, IGF1R, and BRAF inhibitors, for example effectively inhibited cell growth in 10 of the 17 HCC lines, whereas MEK, cell-cycle related kinases and MTOR inhibitors were generally active, effectively suppressing cell growth in 10 of the 17 cell lines.

Conclusion

The Area Under the dose-response Curve (AUC) is a measure of pharmacological efficacy that is used to assess inhibitor responses. Dr. Timothy J. Martins and James Annis of the Quellos HTS Core at the University of Washington's ISCRM supplied us with kinase inhibitor HTS services, which we really appreciate. John D Scott, Anthony K. Leung, David Shechner, and members of the Ong and Maly laboratories provided valuable feedback on the text. Grants R01GM086858, R01GM129090, R01AR065459, R21EB018384, R21CA177402, and K22CA201229-01 from the National Institutes of Health were used to support this study. Using the area under the dose-response curve (AUC) as a pharmacological efficacy metric, we found diverse responses to inhibitors. For example, inhibitors of FGFR,

EGFR, IGF1R, and BRAF successfully stopped cell proliferation in 10 HCC lines, but inhibitors of MEK, cell-cycle inhibitors, and cell-cycle inhibitors did not. The American Cancer Society (133870-RSG-19-197-01-CDD); the Sidney Kimmel Foundation; and a German Research Foundation (DFG) postdoctoral research grant (GO 2358/1-1) were all active, effectively decreasing cell proliferation in 10 HCC lines. The Fibrolamellar Cancer Foundation and the Department of Defense have generously supported us under grant number CA150370. This work made use of an EASY-nLC1200 UHPLC and a Thermo Scientific Orbitrap Fusion Lumos Tribrid mass spectrometer, all of which were purchased with monies through a National Institutes of Health SIG The information is solely the responsibility of the authors and does not necessarily represent the views of the National Institutes of Health. Dr. Timothy J. Martins and James Annis of the Quellos HTS Core at the University of Washington's ISCRM provided us with kinase inhibitor HTS.

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