Cell Cycle

ISSN: 1538-4101 (Print) 1551-4005 (Online) Journal homepage: http://www.tandfonline.com/loi/kccy20

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To cite this article: Fei Chen & Vince Castranova (2009) Beyond apoptosis of JNK1 in liver cancer, Cell Cycle, 8:8, 1145-1147, DOI: 10.4161/cc.8.8.8200

To link to this article: http://dx.doi.org/10.4161/cc.8.8.8200

[Cell Cycle 8:8, 1145-1147; 15 April 2009]; ©2009 Landes Bioscience

Perspective

Beyond apoptosis of JNK1 in liver cancer

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Key words: JNK1, HCC, apoptosis, epigenetics

Hepatocellular Carcinoma (HCC) is the fourth most common neoplasm and the third leading cause of cancer-related death worldwide. Tremendous effort has been made during the past several years in understanding the molecular mechanisms governing the pathogenesis and progression of HCC. Recent studies indicated that c-Jun N-terminal kinase 1 (JNK1), but not JNK2, played pivotal role in the expression of the key signature genes and the prognostic outcomes of HCC. Accordingly, we believe that targeting JNK1 is not only mechanistically sound but also clinically feasible for the treatment of HCC.

It appears to be unequivocal that c-Jun N-terminal kinase (JNK) is a key player in cellular apoptotic responses trigged by a wide spectrum of stress signals.[1] However, there is no shortage of challenges for the pro-apoptotic function of JNK under a considerable number of circumstances.[2] Emerging evidence indicates that JNK is a pivotal kinase providing the cells with a growth advantage during cancerous transformation. To reconcile this profound dilemma, many researchers believe that the opposite function of JNK is cell context dependent.[3] A transient activation of JNK is pro-apoptotic, whereas sustained activation of JNK is anti-apoptotic and consequently, tumorigenic. The situation is further complicated by the existence of three JNK family members, JNK1, JNK2 and JNK3, each of which has several different alternatively spliced isoforms.[4,5] It is conceivable, thus, that each isoform has its own preferential upstream regulators or substrates for interacting, docking and phosphorylation.

Most recently, two groups simultaneously reported that JNK1, but not JNK2, is over-activated in more than 50% of the human hepatocellular carcinoma (HCC) samples in their independent studies.[6,7] Through an immunoprecipitation-based kinase activity assay, Hui et al.[6] demonstrated a substantial enhancement of JNK1 activity in 4 out of 7 HCC samples relative to the case-matched noncancerous liver tissues. Supporting this observation, immunohistochemistry study revealed that about 56% of HCC tissue slides exhibited higher phosphorylated-JNK (pJNK) signal as compared to noncancerous liver tissues. The authors concluded that this enhancement in pJNK is largely attributible to JNK1 but not JNK2, since pJNK2 specific ELISA assay showed roughly equal activity of pJNK2 between HCC and noncancerous liver tissues. A strikingly similar observation was made by Chang et al.[7] who showed that JNK1, but not JNK2, was highly activated in about 55% of human HCC samples. Both teams tried very hard to understand how JNK1 activation is associated with the initiation or progress of HCC. Through an elegant experimental design using JNK1 or JNK2 gene knockout mice, Hui et al.[6] observed a remarkable reduction in HCC size, number and inducibility by DEN in JNK1$^{-/-}$, but not in JNK2$^{-/-}$ mice. Mechanistic studies using mouse liver tissues further indicated that deficiency in JNK1 gene potentiated expression of p21, possibly due to the reduced expression of c-Myc, a well-known oncogene that had been suggested as a transcriptional repressor for p21 in earlier reports[8] (Fig. 1). Interestingly, the previously suspected cronies of JNK1 in the mouse HCC model, such as IL-6,[9] IL-1β,[10] TNFα[11] and several growth factors were not changed between JNK1 knockout and control mice.

A different approach was pursued by Chang et al.[7] in delineating how JNK1 contributes to the development of human HCC by defining signature genes closely linked to higher or lower JNK1 activity in HCC tissue. Not surprisingly, the genes controlling the cell cycle, proliferation and metabolism were overexpressed in the high JNK1 HCC tissue, whereas genes for differentiation, morphogenesis, antioxidant response and p450 family were substantially underexpressed in the high JNK1 HCC tissue relative to the low JNK1 HCC tissue. A surprising observation is the positive association of a number of genes encoding histone methyl transferases with the JNK1 activation status in HCC tissue. These histone methyl transferases include EZH2 and SUV39H2 for tri-methylation of lysine 9 and 27 on histone H3 (H3K9me3 and H3K27me3); MLL3, SMYD3 and SMYD5 for tri-methylation of lysine 4 of histone H3 (H3K4me3), and some other potential methyl transferases (Fig. 1). Indeed, their immunoblotting using tissue lysates from both HCC and the case-matched noncancerous liver tissues showed a straightforward association of H3K4me3 and H3K9me3 with HCC tissues exhibiting high JNK1 activation.

As mentioned earlier, the pro-apoptotic function of the JNK family kinases has been dominating the scientific literature for

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Submitted: 01/29/09; Accepted: 02/16/09

Previously published online as a *Cell Cycle* E-publication: http://www.landesbioscience.com/journals/cc/article/8200

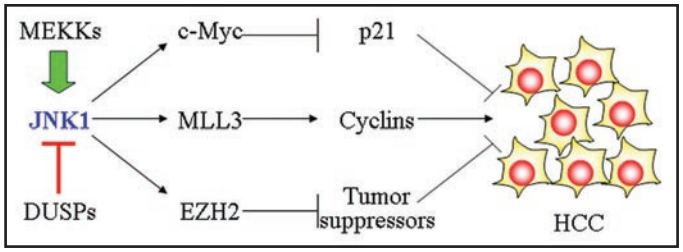

Figure 1. Possible mechanisms of JNK1 activation in HCC development. JNK1 is over-activated in HCC tissue either due to aberrant activation of the upstream kinases, such as MEKKs, or the inactivation of the dual-specific phosphatases (DUSPs). Activated JNK1 upregulates oncogene c-Myc, which represses p21 transcription and causes a faster cell cycle transition of the cells with damaged DNA. JNK1 might be also responsible for the upregulation of MLL3 and EZH2, two histone H3 methyl transferases for the formation of active chromatin marker (H3K4me3) and the silent chromatin markers (H3K9me3 and H3K27me3). The former enhances expression of the cell cycle regulatory genes, such as cyclins, and the latter will silence the transcription of tumor suppressors. All of these pathways will finally facilitate the tumorigenic transformation of hepatocytes which favors development of HCC.

decades.[1] The tumorigenic effect of JNK1 in HCC tissue appears to be contradictory to the universally accepted dogma that JNK kinases enhance cell apoptosis. To tackle such controversy, Hui et al.[6] treated wild type mice with a JNK1 inhibitor followed by DEN treatment for 24 hours and found an appreciable decrease of liver cell apoptosis, implying that the activated JNK1 does induce apoptosis in the early stage of HCC development. This result possibly supports the previous hypothesis that apoptosis, such as under a condition of IKKβ or IKKγ/NEMO deficiency,[12,13] might trigger an over-compensatory proliferation of liver cells to form HCC. The question remaining to be answered, however, is does JNK1 activation foster apoptosis of liver tissue in the middle and later stages, since human HCC tissue with aberrant JNK1 activation is found in later stages. It is not unlikely that the pro-apoptotic effect of JNK1 can be offset or antagonized by the anti-apoptotic and tumorigenic capacity of the JNK1 in HCC tissue at these stages. An additional scenario is that JNK1 acts differently in the earlier stage and later stage of HCC development, because of the overwhelming accumulation of altered biochemical, genetic and epigenetic signals in the later stage of HCC. Without a doubt, these altered signals should be able to induce dramatic effects on the JNK1 signaling network by either aberrantly enforcing activation signals or removing the brakes (negative regulators) for JNK1.

Despite conclusive demonstration indicating a pivotal role of JNK1 in HCC, these two latest studies fell short of offering proof showing that JNK1 activation is indeed causative for HCC progression.[6,7] Neither study explained how JNK1 was over-activated in HCC tissue. Like other MAP kinases, the activity of the JNK1 can be regulated by both upstream kinases and a number of MAP kinase phosphatases, most likely, those dual-specific threonine and tyrosine phosphatases.[14] Is the sustained JNK1 activation in human HCC tissue a result of over-activation of the upstream kinases, or the incapacitation of the dual-specific phosphatases? Genomic mutation of either the kinase genes or the phosphatase genes is rare. In gene expression profiling studies of human HCC tissue, Chang et al.[7] showed an 8- and 4-fold decrease of the DUSP1 and DUSP3, respectively, both of which are able to inactivate JNK1. It is unclear how these JNK1 inhibitors are downregulated in HCC tissue. An additional uncertainty is the role of JNK1 in the opposite expression of the oncogene c-Myc and the tumor suppressor p21. The influence of JNK1 on the upregulation of c-Myc and downregulation of p21 was beautifully revealed in DEN-induced mouse HCC tissue by Hui et al.[6] In contrast, the expression of both c-Myc and p21 appears to be unchanged between high JNK1 HCC tissue and the low JNK1 HCC tissue in studies conducted by Chang et al.[7] Such discrepancy could stem from differences between human and mouse HCCs and the difficulty in aligning tumor stages between these two species.

It is absolutely true that additional studies are much needed to determine the role of JNK1 in the initiation, maintenance and progression of human HCC. If JNK1 is indeed a factor for human HCC development, a new molecular targeting strategy can be evolved for the treatment of this deadly disease that has plagued people in East Asia, South Asia and sub-Sahara Africa for several decades.[15] It is known that tumor cells or pre-cancerous cells in HCC tissue are riddled with numerous genetic errors and epigenetic abnormalities. The changes in gene expression dynamics may be catastrophic in these cells. Although it is highly desirable that molecular targeting can be potentially achieved by identifying which key gene is overexpressed, it appears to be impractical at the present because there are too many genes whose expression is changed in HCC tissue. Furthermore, there is no clear picture of which gene or gene sets are truly molecularly targetable. In contrast, targeting a specific cellular signaling pathway, such as JNK1, is easily manageable. Encouraging data in this regard were presented by Hui et al.[6] who showed that a peptidic JNK inhibitor is profoundly effective in reducing the size and number of HCC lesions induced by DEN in mice. Thus, JNK1 targeting in human HCC therapy is clinically feasible and could be a true milestone in HCC treatment.

Acknowledgements

F.C. is supported by an intramural research grant from the National Institute for Occupational Safety and Health, Centers for Disease Control and Prevention of the USA (970036). We thank the members in F.C.'s laboratory and the Pathology and Physiology Research Branch for critical reading of this manuscript.

References

1. Dhanasekaran DN, Reddy EP. JNK signaling in apoptosis. Oncogene 2008; 27:6245-51.
2. Bode AM, Dong Z. The functional contrariety of JNK. Mol Carcinog 2007; 46:591-8.
3. Davis RJ. Signal transduction by the JNK group of MAP kinases. Cell 2000; 103:239-52.
4. Karin M, Gallagher E. From JNK to pay dirt: jun kinases, their biochemistry, physiology and clinical importance. IUBMB Life 2005; 57:283-95.
5. Chen F. MAPK8 (mitogen-activated protein kinase 8). Atlas Genet Cytogenet Oncol Haematol 2003; URL::http://AtlasGeneticsOncology.org/Genes/JNK1ID196.html.
6. Hui L, Zatloukal K, Scheuch H, Stepniak E, Wagner EF. Proliferation of human HCC cells and chemically induced mouse liver cancers requires JNK1-dependent p21 downregulation. J Clin Invest 2008; 118:3943-53.
7. Chang Q, Zhang Y, Beezhold KJ, et al. Sustained JNK1 activation is associated with altered histone H3 methylations in human liver cancer. J Hepatol 2009; 50:323-33.
8. Gartel AL, Ye X, Goufman E, et al. Myc represses the p21(WAF1/CIP1) promoter and interacts with Sp1/Sp3. Proc Natl Acad Sci USA 2001; 98:4510-5.
9. Naugler WE, Sakurai T, Kim S, et al. Gender disparity in liver cancer due to sex differences in MyD88-dependent IL-6 production. Science (New York, NY) 2007; 317:121-4.

10. Wang Y, Kato N, Hoshida Y, et al. Interleukin-1beta gene polymorphisms associated with hepatocellular carcinoma in hepatitis C virus infection. Hepatology 2003; 37:65-71.
11. Migita K, Miyazoe S, Maeda Y, et al. Cytokine gene polymorphisms in Japanese patients with hepatitis B virus infection—association between TGFbeta1 polymorphisms and hepatocellular carcinoma. J Hepatol 2005; 42:505-10.
12. Maeda S, Kamata H, Luo JL, Leffert H, Karin M. IKKbeta couples hepatocyte death to cytokine-driven compensatory proliferation that promotes chemical hepatocarcinogenesis. Cell 2005; 121:977-90.
13. Luedde T, Beraza N, Kotsikoris V, et al. Deletion of NEMO/IKKgamma in liver parenchymal cells causes steatohepatitis and hepatocellular carcinoma. Cancer Cell 2007; 11:119-32.
14. Rahmouni S, Cerignoli F, Alonso A, et al. Loss of the VHR dual-specific phosphatase causes cell cycle arrest and senescence. Nat Cell Biol 2006; 8:524-31.
15. Llovet JM, Bruix J. Molecular targeted therapies in hepatocellular carcinoma. Hepatology 2008; 48:1312-27.