How transient becomes stable: An epigenetic switch linking liver inflammation and tumorigenesis

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COMMENTARY ON:


Abstract: Hepatocyte nuclear factor 4α (HNF4α) is essential for liver development and hepatocyte function. Here, we show that transient inhibition of HNF4α initiates hepatocellular transformation through a miRNA-inflammatory feedback loop circuit consisting of miR-124, IL6R, STAT3, miR-24, and miR-629. Moreover, we show that, once this circuit is activated, it maintains suppression of HNF4α and sustains oncogenesis. Systemic administration of miR-124, which modulates inflammatory signaling, prevents and suppresses hepatocellular carcinogenesis by inducing tumor-specific apoptosis without toxic side effects. As we also show that this HNF4α circuit is perturbed in human hepatocellular carcinomas, our data raise the possibility that manipulation of this microRNA feedback-inflammatory loop has therapeutic potential for treating liver cancer.

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Hepatocellular carcinoma (HCC) is the most frequent primary tumor of the liver and a leading cause of cancer-related death. This tumor stems from terminally differentiated hepatocytes, or in some cases from resident hepatic progenitor cells. The multistep process of HCC development is initiated by various risk factors, including chronic infection with hepatitis viruses, alcohol, aflatoxin B1, diabetes and obesity. Whatever the etiology, tumorigenesis invariably proceeds through inflammation, liver damage and repeated cycles of apoptosis and regeneration, leading to the accumulation of genetic and epigenetic alterations [1] (Fig. 1).

Using rodent models of liver disease, key intracellular signal transducers linking liver inflammation to HCC have been unraveled. Recent focus on NF-κB has revealed diverse and apparently opposite functions in hepatic inflammation, fibrosis, and HCC induction. It has been shown that activated IkappaB kinase (IKK)/NF-κB pathway may play a tumor-promoting role by protecting tumor cells from death or enhancing their proliferation [2]. In another model, liver-specific expression of lymphotxin alpha and beta induces liver inflammation and HCC. By contrast, in several studies, deletion of NEMO/IKKgamma caused systemic hepatitis and HCC by inhibiting the antiapoptotic function of NF-κB [3]. This phenotype could be rescued by hepatocyte-specific STAT3 ablation. Moreover, deletion of the MAP3-kinase TGF-beta-activated kinase 1 (TAK1) in liver parenchymal cells could block TNF-induced NF-κB activation and caused hepatocyte dysplasia and HCC.

Importantly, IL6, a target of NF-κB, activates the STAT3 signaling pathway. This pathway has been implicated in the tumor-promoting effect of obesity in murine models of liver tumorigenesis [4], and it might account for gender disparity in HCC incidence both in humans and mice. Thus, modulation of the inflammatory response by the IL6-STAT3 and NF-κB pathways contributes to enhance the risk of HCC development in the injured liver.

While liver inflammation and hepatocyte proliferation are recognized as the driving forces of liver cell transformation, little is known so far on the role of epigenetic changes in the stabilization of the cellular transformed phenotype that gives rise to cancer. An interesting mechanistic model supporting the early involvement of epigenetic players has been recently proposed by the teams of Hadzopoulou-Cladaras and Lliopoulos [5]. This study started with the observation that treatment of immortalized hepatic cell lines with siRNAs against the nuclear hormone receptor HNF4α caused cell-specific transformation. HNF4α was previously shown to suppress HCC development by repressing epithelial-mesenchymal transition, thereby maintaining the epithelial identity of hepatocytes [6]. HNF4α orchestrates a transcription factor network that controls hepatocyte differentiation and liver morphogenesis [7]. At later stages, HNF4α expression is pivotal for the establishment of liver architecture, zonation and functional activity. The broad effect of HNF4α on liver functions has been recently involved in the regulation of cytokine-induced inflammatory responses.
In the study commented here, Hatziapostolou et al. observed that siRNA-induced inhibition of HNF4α expression persisted for 2 months, beyond the predicted duration of silencing by short-lived siRNAs. The hypothesis of a positive feedback loop was suggested by a previous work of the same team, showing the predominant role of miRNAs in a regulatory circuitry linking inflammation to breast cancer [8]. The search for miRNAs that directly target HNF4α led to the identification of miR-24 and miR-629. The finding that ectopic expression of these two miRNAs can transform immortalized hepatocytes as efficiently as the downregulation of HNF4α is probably one of the most striking observations of this work. Such data evoke the activity of oncogenic miRNAs (oncomiRs) and highlight the reciprocal regulation of specific oncomiRs and their tumor suppressor targets.

The second step was the identification of a STAT3 binding site in miR-24 and miR-629 promoters. Binding of STAT3 was validated by chromatin immunoprecipitation, and IL6-mediated STAT3 activation was associated with increased expression of the two miRNAs. Increased STAT3 phosphorylation in cells over-expressing miR-24 and miR-629 indicated the existence of a positive feedback loop. The search for miRNAs that could be regulated by HNF4α and in turn block STAT3 activation resulted in the identification of miR-124 as a direct target of miR-24 (Fig. 1). These quite remarkable data obtained in cell lines were validated in murine HCC models and by analysis of the expression levels of the different protagonists in human HCCs.

The HNF4 inflammatory circuit emphasizes the complexity of miRNA-transcription factor crosstalks in liver inflammation and HCC, as previously outlined in recent studies that pinpointed miRNAs as co-effectors in hepatic inflammatory processes [9]. miR-26 is known to target IL6, and accordingly, low levels of miR-26 in HCC have been correlated with elevated IL6 expression, activated NF-xB pathway, and poor patients' survival [10]. HNF4α controls the expression of miR-122, an abundant liver-specific miRNA with dual roles in stimulating hepatitis C virus translation and in conveying some of the tumor suppressive effects of HNF4α. Notably, a regulatory circuitry involving miR-122, C/EBPα, GSK3-β, and IGF-1R has been implicated in liver carcinogenesis. Moreover, induction of the oncogenic miRNA miR-155 by proinflammatory cytokines and NF-xB has been implicated in alcoholic liver diseases and at early stages of non-alcoholic steatohepatitis (NASH) (reviewed in [9]).

The work by Hatziapostolou et al. provides insights into the ability of transient epigenetic changes to initiate a stable feedback loop implicated in the transition between non-transformed and transformed states. Given the therapeutic potential of manipulating miRNA expression, a major challenge will be to
propersly integrate these insights into a path to the clinic that will be exploited in the treatment of patients with HCC.

Conflict of interest

The authors declared that they do not have anything to disclose regarding funding or conflict of interest with respect to this manuscript.

References
