

Novel Biomarkers with Potential Utility in Diagnosis of liver Fibrosis

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ABSTRACT

Liver fibrosis, which is a consequence of chronic liver diseases, is associated with excessive deposition of extracellular matrix (ECM) proteins during the dynamic process of repair and regeneration. Hence the events associated with repair and regeneration could be explored as predictable markers to assess liver fibrosis. We report here the possible utility of a few biomarkers in early diagnosis of liver fibrosis.

Key words: Liver fibrosis, Biomarker, Chronic liver disease, Non-invasive diagnosis, Diagnostic imaging.

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DOI: 10.5530/BEMS.1.2.3

INTRODUCTION

There are four major stages of fibrogenic response,¹⁻³ i.e., 1) initiation of the response (triggered by the primary cause of liver injury), 2) activation of effector cells, 3) elaboration of extracellular matrix (ECM), 4) Dynamic and progressive deposition of ECM leading to liver failure. All these four stages involve a much complex and dynamic interactions of resident cellular architecture (in this case hepatocytes), resident/non-resident progenitor cells, locally released growth factors/cytokines, and systemically released chemokine's. Fibrosis is not unique to liver and neither are the various cellular and biochemical pathways involved in fibrosis, thus making the discovery and development of biomarkers difficult. Cells such as fibroblasts and cytokines such as transforming growth factor beta (TGF-beta) are core elements to various stages of fibrosis (Figure 1). Recently several other cell types are implicated in inflammation driven fibrosis response, some of which are myeloid cells, mesenchymal cells and various types of macrophages, all of which primarily trigger excess extracellular matrix production.^{2,3}

Incidentally most fibrogenic myofibroblasts (MFB) are endogenous to the liver, coming from hepatic stellate cells (HSC) and portal fibroblasts. Dysregulated inflammatory responses have been associated with most (if not all) hepatotoxic insults and chronic oxidative stress, play a role during the initial liver inflammatory phase and its progression to fibrosis. Redox-regulated processes¹ are responsible for activation of HSC to MFB, as well as maintenance of the MFB function. Increased oxidative stress also induces hepatocyte apoptosis, which contributes to increase the liver injury and to transdifferentiate HSC to MFB, favouring the fibrogenic process.

FGFs are multifunctional proteins with regulatory, morphological, and endocrinal effects and are involved in angiogenesis, wound healing, and embryonic development. In humans, 22 members of the FGF family have been identified, all of which are structurally related signaling molecules. FGFR4 is the predominant isoform of the FGFRs in hepatocytes and may have a major role in progression of liver fibrosis.

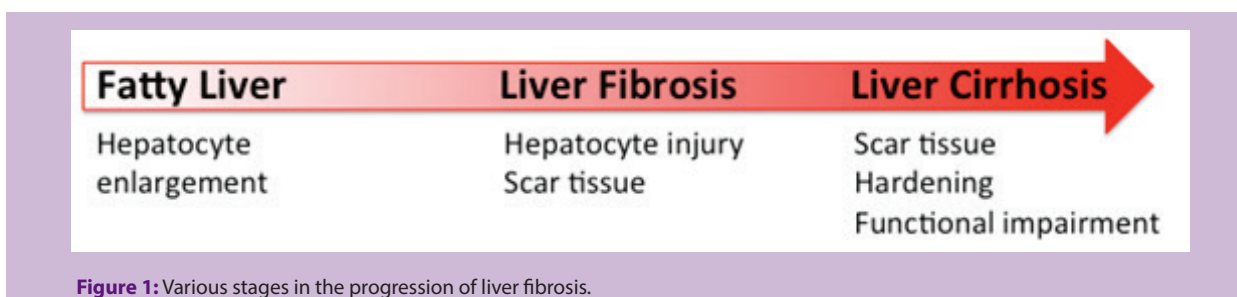
High risk of liver cirrhosis and markedly increase in the alpha fetoprotein AFP level (> 4 ug/ml) are reported in patients with Hepato Cellular Carcinoma.

High levels of Bone morphogenetic protein (BMP) in fibrosis has been reported.

Based on the above facts I suggest the following markers, which may be useful in diagnosis of liver fibrosis.

Contrast perfusion index (Ultrasound imaging)

While ultrasound imaging is routinely used to image liver, a contrast based imaging may significantly improve the assessment of liver fibrosis. As fibrosed part is less vascular compared to health liver tissue, we will see a difference in the perfusion index of healthy Vs fibrosed liver tissue. Which may be represented as a perfusion ratio, i.e., Ratio of contrast perfusion in fibrosed Vs Healthy liver tissue. A Higher ratio will be reflecting progressive fibrosis while a lower ratio will suggest regression of fibrosis. Hence this approach will be helpful in both assessing the pathology and as well as impact of any therapeutic intervention. The perfusion ratio based approach can also be applied to other imaging modalities such as CT or MRI where available.



A few contrasts for ultrasound imaging are commercially available. These contrast agents can be administered intravenously in association with ultrasound imaging to estimate the contrast perfusion index. The procedure will be quick and can be performed within 15-20 min. Software program can be developed to automate the analysis on the ultrasound imaging devices.

FGF23/IGF1 ratio

As mentioned in the introduction section above fibroblasts and FGF are implicated in several tissue fibrosis process. Although I specifically pinpoint this on FGF23 due to recent reports on this isoform, the other relevant FGF may also be looked into in patients with different version of liver fibrosis.

IGF1 is a very vital growth factor involved in cell proliferation and specifically its anti-apoptotic effect is of considerable therapeutic utility. Liver is a major source of IGF1 while macrophages are major extra hepatic source of IGF1. Since liver fibrosis process (fatty liver) begins with hepatocyte enlargement (Figure 1), it is likely IGF1 has a role in this stage and likewise the macrophages, which drive the inflammatory process associated with the liver fibrosis, also contribute to IGF1 in circulation.

Hence FGF23/IGF1 ratio may reflect a dynamic process during liver fibrosis involving hepatocyte hypertrophy, scar tissue formation and inflammatory macrophage activity.

Increase in FGF23/ IGF1 ratio may indicate progressive liver fibrosis.

This index must be in association with currently use liver specific pathology markers such as ALT/AST ratio and AST/Platelet ratio. This is necessary as both FGF23 and IGF1 may not be specific to liver but will be specific to fibrosis events.

Commercially available ELISA kits can be used for FGF23 and IGF1 detection in serum samples. It may take up to 48 hrs to perform this test in any laboratory setting equipped to perform ELISA analysis.

CD34, Cx3Cr1 and cytokeratin-7 positive cells in circulation

CD34, Cx3Cr1 and cytokeratin-7 have been implicated in scarring, fibrosis and inflammation. Hence it is common to observe cells positive

for these markers in circulation following inflammation and tissue fibrosis. It is likely that cytological profile of these markers in blood may be associated with liver fibrosis. Like FGF23 and IGF1, these cytological markers may not be specific to liver pathology, however they may be correlated with ALT/AST ratio and/or AST/Platelet ratio to assess any change in the cytological profile of CD34, Cx3Cr1 and cytokeratin-7 in circulation with liver pathology.

Cytological profiling of CD34, Cx3Cr1 and cytokeratin-7 will require a flow cytometer, however this technology is getting affordable. Cytological profiling process itself is quick (less than 4 hrs). In the cytological profiling, CD34, Cx3Cr1 and cytokeratin-7 positive cells may be assessed independently or in combination. High levels of CD34 and Cx3Cr1, Cx3Cr1 and cytokeratin-7, CD34 and cytokeratin-7, or CD34, Cx3Cr1 and cytokeratin-7 positive cells may be an index of progressive liver fibrosis.

CONCLUSION

We report here the possible use of some novel biomarkers in diagnosis of liver fibrosis.

ACKNOWLEDGEMENT

Support from University College Dublin-Seed funding (AHSK) and Stemology (AHSK) is acknowledged.

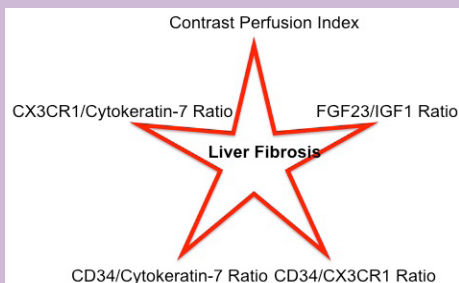
CONFLICT OF INTEREST

The author declare no conflict of interest.

REFERENCES

1. Crosas-Molist E, Fabregat I. Role of NADPH oxidases in the redox biology of liver fibrosis. *Redox Biol.* 2015;14(6):106-11.
2. Yan Z, Qu K, Zhang J, Huang Q, Qu P, Xu X, et al. CD147 promotes liver fibrosis progression via VEGF-A/VEGFR2 signalling-mediated cross-talk between hepatocytes and sinusoidal endothelial cells. *Clin Sci (Lond).* 2015;129(8):699-710.
3. Kamada Y, Ono M, Hyogo H, Fujii H, Sumida Y, Mori K, et al. A novel noninvasive diagnostic method for nonalcoholic steatohepatitis using two glyco-biomarkers. *Hepatology.* 2015. doi: 10.1002/hep.28002.

PICTORIAL ABSTRACT



Dr Kumar: Is a Clinician, Drug Discovery Scientist, Entrepreneur and Science journalist with over 15 years of research and teaching experience in pharmacology and regenerative medicine. He has Extensive international experience with over 80 peer-reviewed publications. He has attracted investments from several national and international organisations at various stages of his carrier. He has successfully directed several projects in preclinical /clinical pharmacology, specifically in cardiovascular pathophysiology, diabetic complications, medical devices, arthritis, and regenerative medicine, which has resulted in either patentable products and/or high impact publications.

SUMMARY

- Contrast perfusion index (Ultrasound imaging).
- FGF23/IGF1 ratio.
- Circulating levels of CD34, Cx3Cr1 and cytokeratin-7 positive cells.

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