

- [3] Holmström P, Gafvels M, Eriksson LC, Dzikaite V, Hultcrantz R, Eggertsen G, et al. Expression of iron regulatory genes in a rat model of hepatocellular carcinoma. *Liver Int* 2006;26:976–985.
- [4] Youn P, Kim S, Ahn JH, Kim Y, Park JD, Ryu DY. Regulation of iron metabolism-related genes in diethylnitrosamine-induced mouse liver tumors. *Toxicol Lett* 2009;184:151–158.
- [5] Roessler S, Jia H-L, Budhu A, Forgues M, Ye Q-H, Lee J-S, et al. A unique metastasis gene signature enables prediction of tumor relapse in early-stage hepatocellular carcinoma patients. *Cancer Res* 2010;70:10202–10212.
- [6] Kijima H, Sawada T, Tomosugi N, Kubota K. Expression of hepcidin mRNA is uniformly suppressed in hepatocellular carcinoma. *BMC Cancer* 2008;8:167–175.
- [7] Girelli D, Pasino M, Goodnough JB, Nemeth E, Guido M, Castagna A, et al. Reduced serum hepcidin levels in patients with chronic hepatitis C. *J Hepatol* 2009;51:845–852.
- [8] Miura K, Taura K, Kodama Y, Schnabl B, Brenner DA. Hepatitis C virus-induced oxidative stress suppresses hepcidin expression through increased histone deacetylase activity. *Hepatology* 2008;48:1420–1429.
- [9] Weizer-Stern O, Adamsky K, Margalit O, Ashur-Fabian O, Givol D, Amariglio N, et al. Hepcidin, a key regulator of iron metabolism, is transcriptionally activated by p53. *Br J Haematol* 2007;138:253–262.
- [10] Nault J-C, Zucman-Rossi J. Genetics of hepatocellular carcinoma: the next generation. *J Hepatol* 2014;60:224–226.

Sonja M. Kessler^{*,†}

Stephan Laggai

Alexandra K. Kiemer

Department of Pharmacy, Pharmaceutical Biology,
Saarland University, 66123 Saarbrücken, Germany

*Corresponding author.

E-mail address: s.kessler@mx.uni-saarland.deAhmad Barghash[†]

Volkhard Helms

Center for Bioinformatics, Saarland University,
66123 Saarbrücken, Germany

† These authors contributed equally to this work.



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Reply to: “Hepatic hepcidin expression is decreased in cirrhosis and HCC”

Hepcidin in chronic liver disease and hepatocellular carcinoma: The plot thickens

To the Editor:

We read with interest the letter by Kessler *et al.* [1] that demonstrated decreased hepcidin levels in various stress situations as well as in hepatocellular carcinoma. Lowered hepcidin levels seen in diethylnitrosamine-treated mice nicely complement earlier data on mice subjected to thioacetamide-induced liver fibrosis or administered Lieber-DeCarli diet (an experimental model of alcoholic liver disease), and suggest that diminished hepcidin levels represent a common reaction to various liver stresses [1–3]. Similarly, observations predominantly made in hepatitis B-infected patients extend previous findings, obtained in subjects with chronic liver disease due to hepatitis C infection or excessive alcohol intake, which all display reduced liver hepcidin levels [1,4]. Most importantly, in an elegant molecular analysis, Kessler *et al.* also showed that hepatocellular carcinomas exhibit diminished hepcidin expression, that was previously reported only in smaller studies.

Since hepcidin represents the central negative regulator of iron metabolism [4], these data reinforce the hypothesis that chronic liver disorders may promote development of acquired iron overload again triggering the progression of liver fibrosis and/or development of hepatocellular carcinoma. As an underlying mechanism, iron overload has multiple deleterious downstream effects, such as formation of reactive oxygen species, mitochondrial or lysosomal injury [5,6]. However, while several reports demonstrated an association between increased hepatic iron load and progression of liver fibrosis and/or HCC development [7], no such studies are available for hepcidin. This is rather surprising given the availability of multiple assays that can conveniently assess hepcidin serum levels. However, the extent to

which these assessments can accurately reflect liver hepcidin expression or iron content in complex clinical settings, comprising multiple confounding factors, remains to be clarified. Nevertheless and without any doubt, such studies are of obvious medical and biological interest, although their interpretation will likely be complicated.

In that respect, the regulation of hepcidin production is complex and is affected, not only by iron metabolism, but also by various hepatic factors; by inflammation, erythropoietic drive, hypoxia etc. [4]. Moreover, hepcidin has a short half-life and displays a circadian rhythm [8]. Even more challenging will be to dissect whether the altered hepcidin levels represent a cause or consequence of liver disease/fibrosis progression. Finally, the deleterious effects of iron metabolism in liver carcinogenesis are further modulated by complex genetic factors at both the constitutional and functional levels; indeed, multiple genetic traits seem to impact hepatic iron content in patients with chronic liver disease [9]. While in-depth transcriptomic analyses reveal that expression of the *HAMP* gene could participate in a refinement of molecular classification of HCCs [10].

In conclusion, although the road ahead will likely be bumpy, it will be challenging to uncover the complex interaction between liver disease and iron metabolism. These interactions are now more important than ever given the rapid emergence of hepcidin-targeted therapeutic strategies that could be further modulated by host- and/or tumour-related genetic factors in the setting of personalized medicine.

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

- [1] Kessler Sonja M, Barghash Ahmad, Laggai Stephan, Helms Volkhard, Kiemer Alexandra K. Hepatic hepcidin expression is decreased in cirrhosis and HCC. *J Hepatol* 2015;62:977–979.
- [2] Harrison-Findik DD, Schafer D, Klein E, Timchenko NA, Kulaksiz H, Clemens D, et al. Alcohol metabolism-mediated oxidative stress down-regulates hepcidin transcription and leads to increased duodenal iron transporter expression. *J Biol Chem* 2006;281:22974–22982.
- [3] Mueller K, Sunami Y, Stuetzle M, Guldiken N, Kucukoglu O, Mueller S, et al. CHOP-mediated hepcidin suppression modulates hepatic iron load. *J Pathol* 2013;231:532–542.
- [4] Meynard D, Babitt JL, Lin HY. The liver: conductor of systemic iron balance. *Blood* 2014;123:168–176.
- [5] Ramm GA, Ruddell RG. Iron homeostasis, hepatocellular injury, and fibrogenesis in hemochromatosis: the role of inflammation in a noninflammatory liver disease. *Semin Liver Dis* 2010;30:271–287.
- [6] Lunova M, Goehring C, Kuscuoglu D, Mueller K, Chen Y, Walther P, et al. Hepcidin knockout mice fed with iron-rich diet develop chronic liver injury and liver fibrosis due to lysosomal iron overload. *J Hepatol* 2014;61:633–641.
- [7] Valenti L, Fracanzani AL, Bugianesi E, Dongiovanni P, Galmozzi E, Vanni E, et al. HFE genotype, parenchymal iron accumulation, and liver fibrosis in patients with nonalcoholic fatty liver disease. *Gastroenterology* 2010;138:905–912.
- [8] Ganz T. Iron homeostasis: fitting the puzzle pieces together. *Cell Metab* 2008;7:288–290.
- [9] Nahon P, Sutton A, Rufat P, Charnaux N, Mansouri A, Moreau R, et al. A variant in myeloperoxidase promoter hastens the emergence of hepatocellular carcinoma in patients with HCV-related cirrhosis. *J Hepatol* 2012;56:426–432.
- [10] Boyault S, Rickman DS, de Reyniès A, Balabaud C, Rebouissou S, Jeannot E, et al. Transcriptome classification of HCC is related to gene alterations and to new therapeutic targets. *Hepatology* 2007;45:42–52.

Mariia Lunova
Department of Medicine III and IZKF,
University Hospital Aachen, Germany
Institute for Clinical and Experimental Medicine,
Prague, Czech Republic

Christian Trautwein
Pavel Strnad*
Department of Medicine III and IZKF,
University Hospital Aachen, Germany
*Corresponding author.
E-mail address: pstrnad@ukaachen.de

Pierre Nahon
APHP, Jean Verdier Hospital, Bondy, France
Paris 13 University, Bobigny, France
INSERM U1162, Génomique fonctionnelle des Tumeurs solides,
Paris, France



HCC and liver disease risks in homozygous *PNPLA3* p.I148M carriers approach monogenic inheritance

To the Editor:

The study published in the *Journal of Hepatology* by Liu *et al.* [1] has robustly validated a strong genetic association between the common *PNPLA3* variant p.I148M and the risk of developing hepatocellular carcinoma (HCC), in patients with non-alcoholic fatty liver disease (NAFLD), also reported previously [2]. The association study [1] comprised 100 individuals with NAFLD-associated HCC and 1,476 controls and showed that homozygous carriers of the p.I148M mutation carry a 12-fold increased HCC risk (odds ratio [OR] = 12.19, 95% confidence interval = 6.89–21.58, $p < 0.0001$) as compared to p.I148 homozygotes. The magnitude of this risk is remarkable and comparable to another inherited liver disease, i.e., hereditary haemochromatosis. This disease is due to *HFE* gene mutations and it represents a monogenic ('Mendelian') liver disorder [3].

Table 1A shows that the OR for HCC development is comparable between *HFE*-associated haemochromatosis and *PNPLA3*-associated fatty liver disease. A meta-analysis on haemochromatosis, encompassing in total 320 HCC cases and almost 2,000 controls from Europe and China [4], showed that homozygous carriers of the *HFE* mutation p.C282Y have a 12-fold increased risk of developing liver disease (Table 1A). Interestingly, carriers of *HFE* and *PNPLA3* mutations may develop HCC even without cirrhosis [5,6].

Overall, both *PNPLA3* p.I148M and *HFE* p.C282Y mutations pose comparable risks for developing liver disease. Interestingly,

studies showed the *PNPLA3* p.I148M allele has been associated with a 2- to 7-fold increased risk for fatty liver, steatohepatitis, and fibrosis [7,8]. This effect magnitude is comparable to the 4- to 10-fold increased risk of liver disease and steatohepatitis, respectively, in homozygous carriers of the *HFE* mutation [4] (Table 1B). Similarly, the interaction between the genetic variant and alcohol consumption promotes the development of clinically relevant liver disease for both *PNPLA3* and *HFE* mutations [9,10]. The frequency of the *PNPLA3* p.I148M mutation in the general population is higher than the frequency of the *HFE* p.C282Y mutation (Table 1B). This results in an *a priori* higher chance of carrying the *PNPLA3* rather than *HFE* variant in NAFLD-associated HCC.

However, whereas *HFE*-associated haemochromatosis represents a well-recognized monogenic disease, *PNPLA3*-associated steatohepatitis currently does not. We conclude that given the remarkably high risk of HCC secondary to NAFLD for *PNPLA3* p.I148M homozygotes, there is a need for additional systematic studies with long-term follow-up. Finally, well-powered genetic studies to investigate the risk of HCC conferred by combinations of *HFE* and *PNPLA3* mutations might be envisioned.

Conflict of interest

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