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Abstract: Whether the epithelial cell response (named "ductular reaction") observed in chronic situations is only a marker of severity of liver disease or plays a role in liver cell regeneration remains under debate. However, recent cell tracking experiments provide a robust argument for the differentiation of those cells in an animal model of chronic liver disease and indicate that the situation could be similar in humans (Deng et al. 2018). Thanks to three other human studies (Lin et al., 2010; Yoon et al. 2011; Lanthier et al. 2015), we believe that epithelial cells give rise to subsequent peribiliary intermediate hepatocytes and create fully functional adjacent hepatocytes that may be beneficial for human liver regeneration.

Cover letter

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Resident liver progenitor cells:

proofs of their contribution to human liver regeneration

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Abstract

Whether the epithelial cell response (named “ductular reaction”) observed in chronic situations is only a marker of severity of liver disease or plays a role in liver cell regeneration remains under debate. However, recent cell tracking experiments provide a robust argument for the differentiation of those cells in an animal model of chronic liver disease and indicate that the situation could be similar in humans (Deng et al. 2018). Thanks to three other human studies (Lin et al., 2010; Yoon et al. 2011; Lanthier et al. 2015), we believe that epithelial cells give rise to subsequent peribiliary intermediate hepatocytes and create fully functional adjacent hepatocytes that may be beneficial for human liver regeneration.

Under normal conditions, mature hepatocytes are able to proliferate in order to provide physiological turnover. In acute severe liver injuries and in chronic decompensated situations, this proliferation activity of background hepatocytes could be impaired. In those situations, invasion of the liver by resident epithelial cells occurs. Those cells, located in basal conditions in the canals of Hering which are the proximal branches of the biliary tree, and expressing epithelial cell markers such as keratin 7 or 19, are often named liver progenitor cells (LPC). However, many data support the idea that this epithelial cell response (the ductular reaction) is only a marker of severity of liver disease, could even be detrimental and does not play any role in liver cell regeneration. According to some other data, cells in the ductular reaction may represent dedifferentiated injured mature hepatocytes expressing epithelial markers rather than being LPC [1].

Indeed, experiments in animal models using acute or repeated single injuries did not demonstrate any massive repopulation of the liver by those cells. However, recently, Xing Deng and his colleagues provide interesting data by cell tracking experiments showing that those biliary epithelial cells differentiate into mature hepatocytes in chronic situations mimicking human chronic liver diseases. Interestingly, the authors also analyze human biopsy specimens from patients with chronic liver diseases and evidence the same bi-phenotypic cells (with markers from both LPC and hepatocytes as in animal cell tracking pictures) as a feature of human cirrhosis [2].

Those data provide a robust argument for the differentiation of LPC in an animal model of chronic liver disease and indicate that the situation could be similar in humans. As liver epithelial cell tracking experiments are not technically feasible in humans, we want here to get back on two indirect available proofs showing that LPC

are able in humans to give rise to new hepatocytes [3, 4] and on one other argument suggesting that LPC play a beneficial role in liver regeneration [5].

First, the group of Wey-Ran Lin and Malcolm Alison studied DNA mitochondrial mutations in human cirrhosis from various etiologies [3]. They used those mutations as clonal markers for lineage tracing in the liver. For the first time, they provide evidence that the majority of hepatocytes from cirrhotic nodules harbored the same mutation(s) as the adjacent surrounded keratin 19 positive LPC from the ductular reaction.

Second, So-Mi Yoon and his colleagues investigated the patterns of expression of Epithelial Cell Adhesion Molecule (EpCAM) in chronic hepatitis B and C patients [4]. EpCAM is a surface marker of LPC that is absent on hepatocytes. With immunohistochemical studies, they confirmed that this EpCAM marker was present on the cytoplasm of cholangiocytes (in particular forming the canals of Hering) in normal livers but also in cells from the ductular reaction in severe disease stages and interestingly in hepatocytes in contiguity with ductular cells. Cirrhotic livers had more than fifty percent of EpCAM positivity and those cells were also characterized by important proliferation, evaluated by the proliferating cell nuclear antigen (PCNA) staining. Using in situ hybridization, they evaluated telomere length and were able to evidence a gradual telomere shortening from ductular reaction to EpCAM positive hepatocytes then to EpCAM negative hepatocytes, supporting that those EpCAM negative hepatocytes could originate from LPC.

Third, in patients with decompensated alcoholic liver disease, we carefully determined the number and morphology of keratin 7 positive cells, which we subsequently correlated to patients' outcome and liver histology repeated after 3 months [5]. We were able to demonstrate that patients with improved liver function

during follow-up exhibited at baseline a particular immunohistochemical pattern with a significant higher number of proliferative keratin 7 positive LPC (double Ki67 proliferative nuclei marker and keratin 7 cells from all cell subtype: isolated small progenitor cells, cells from the ductular reaction and larger intermediate hepatocytes) compared to patients with persistent liver failure (non-improvers). To our knowledge, this is the first evidence that this proliferative subtype LPC compartment is a positive prognostic factor in human chronic liver diseases, helping for the improvement of the hepatocellular function. Interestingly, the number of total keratin 7 positive cells (correlated with disease severity) was the same between the two groups (improvers and non-improvers), supporting the further differentiation of proliferating LPC towards hepatocytes.

Collectively, thanks to the recent study from Xing Deng and to those three studies, we believe that LPC give rise to subsequent peribiliary intermediate hepatocytes and create fully functional adjacent hepatocytes that may be beneficial for human liver regeneration in chronic diseases. Further analyses on the subtype of those beneficial epithelial cells and on the important microenvironment needed for their differentiation will provide new therapeutic options in human liver diseases.

Figure legend:

Model of differentiation of biliary epithelial cells (also called liver progenitor cells) to new hepatocytes in human chronic liver diseases, based on recent experimental data. Brown nuclei depict proliferative cells.

LPC: liver progenitor cells, K: keratin, HNF4α: hepatocyte nuclear factor 4 alpha, EpCAM: epithelial cell adhesion modecule.

References

[1] Lanthier N, Rubbia-Brandt L, Spahr L. Liver progenitor cells and therapeutic potential of stem cells in human chronic liver diseases. Acta Gastroenterol Belg 2013;76:3-9.

[2] Deng X, Zhang X, Li W, Feng RX, Li L, Yi GR, et al. Chronic Liver Injury Induces Conversion of Biliary Epithelial Cells into Hepatocytes. Cell Stem Cell 2018;23:114-122.

[3] Lin WR, Lim SN, McDonald SA, Graham T, Wright VL, Peplow CL, et al. The histogenesis of regenerative nodules in human liver cirrhosis. Hepatology 2010;51:1017-1026.

[4] Yoon SM, Gerasimidou D, Kuwahara R, Hytiroglou P, Yoo JE, Park YN, et al. Epithelial cell adhesion molecule (EpCAM) marks hepatocytes newly derived from stem/progenitor cells in humans. Hepatology 2011;53:964-973.

[5] Lanthier N, Rubbia-Brandt L, Lin-Marq N, Clement S, Frossard JL, Goossens N, et al. Hepatic cell proliferation plays a pivotal role in the prognosis of alcoholic hepatitis. J Hepatol 2015;63:609-621.

Figure

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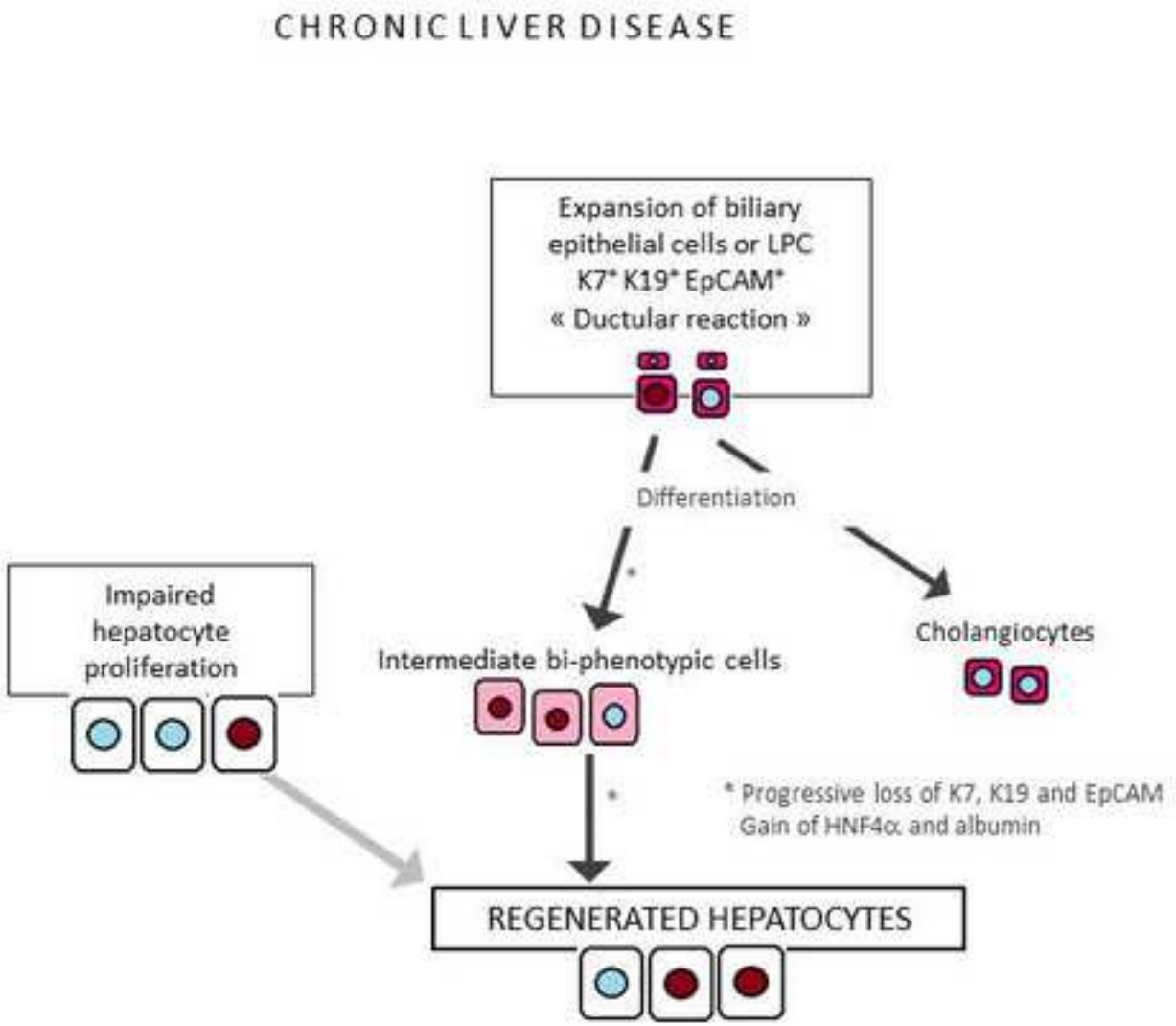

Declaration of interest

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