Editorial

Vascular endothelial growth factors in progenitor cells mediated liver repair

Carlo Spirli[1], Mario Strazzabosco[1,2]

[1]Department of Internal Medicine, Section of Digestive Diseases, Yale University, New Haven, CT, USA; [2]Department of Clinical Medicine, University of Milano-Bicocca, Milan, Italy

Corresponding to: Carlo Spirli, Ph.D, Asst., Professor of Medicine. Yale University, Department of Internal Medicine, Section of Digestive Diseases, 333 Cedar Street-1080 LMP, P.O. Box 208019, New Haven, CT 06520-8019, USA. Email: carlo.spirli@yale.edu.

Submitted Nov 20, 2012. Accepted for publication Dec 20, 2012.
doi: 10.3978/j.issn.2304-3881.2012.12.05
Scan to your mobile device or view this article at: http://www.thehbsn.org/article/view/1322/1846

Hepatic progenitor cells (HPC) are a bipotent cell population of the liver that may differentiate towards hepatocytes or cholangiocytes (1-3). HPC are mostly quiescent in normal livers while proliferate following liver injury. The activation of HPC results in mature hepatocytes or mature cholangiocytes or, in case of chronic damage, in the appearance of "reactive" cholangiocytes (2,3). The latter are small cells positive for cytokeratin 7 and 19 that try to form new ducts but result in clusters that not encircle a well-defined lumen. This reactive process that, in human livers, is mainly localized at the interface between the portal and parenchymal compartments is called ductular reaction" (DR) (4). DR is a well-known reparative mechanism of the liver and several studies have shown how these reparative mechanisms recapitulate developmental liver morphogenetic processes (3,5-8). Reparative processes are different between biliary and hepatocellular diseases, and involve different signaling mechanisms, for example Notch (9-11) or Wnt (12) for biliary or hepatocellular specification, respectively. Reactive cholangiocytes are often confused with hepatic progenitor cells; in fact, the evidence that reactive cholangiocytes are bipotential is scant. In chronic conditions, reactive cholangiocytes correlate with fibrosis and disease progression, indicating that they are the result of pathologic, rather than physiologic repair (3). In fact, "reactive" cholangiocytes re-expresses growth factors, transcription factors and morphogens enabling an active cross-talk between biliary, mesenchymal, vascular and inflammatory cells (3,6). Among these factors, vascular endothelial growth factor (VEGF) and angiopoietins have drawn considerable attention (5,13,14).

VEGF is a complex system of six different factors: VEGF-A, -B, -C, -D and -E and placenta growth factor. Together with its receptors VEGFR1 (Flt-1), VEGFR2 (Flk-1), VEGFR3 (Flt-4) and angiopoietins, VEGF is involved in the regulation of vascular growth, permeability, migration and survival of endothelial cells (15). Although originally thought to be restricted to vascular cells, recent studies have shown that VEGF, together with its receptors, is expressed and functional also in epithelial cells. In particular, in cholangiocytes, VEGF-A appears to regulate VEGF regulate cell proliferation and cross-talk during development, as well as in normal and diseased conditions (5,13,16-18). During liver development, VEGF is a key signal, able to link bile ducts and the network of capillaries emerging from the finest branches of the hepatic artery known as peribiliary plexus (PBP) (13). In fact, the developing bile ducts produce VEGF-A which in turn acts on endothelial cells and their precursor to promote arterial and PBP vasculogenesis (13). Similarly, in ductal plate malformations (DPM), the dysmorphic bile ducts actively secrete VEGF-A and are surrounded by an increased number of vascular structures (19). This is particular evident in cystic cholangiopathies where, in addition to secreting VEGF-A the biliary epithelium expresses VEGFR-2 receptor, that respond to VEGF by increasing proliferation and cyst growth (13). Studies in animal models of Autosomal Dominant Polycystic Kidney Disease (ADPKD) indicate that VEGF stimulates the progression of liver cysts in via autocrine stimulation of cholangiocytes proliferation and paracrine induction of pericystic angiogenesis (17,18). In fact, VEGF induces cell proliferation through the activation of PKA/ERK1/2 signaling, the most important proliferative pathway in cholangiocytes. In turn, an altered cAMP/PKA/

ERK1/2 signaling is responsible of the increased hypoxia-inducible factor 1 α-mediated VEGF secretion (16-18). The blockade of this signaling using inhibitors of VEGFR-2 or mTOR or cAMP production resulted in a significantly decreased in cyst growth (17,18).

In this issue of Hepatobiliary Surgery and Nutrition, Franchitto and colleagues shows that in chronic liver diseases, such as primary biliary cirrhosis (PBC) and HCV-related cirrhosis, VEGF is expressed in HPC and ductular reactive cells (20). In particular, the results show that expansion of HPC is more extensive in PBC with respect to HCV samples. PBC samples were also characterized by a more extensive angiogenesis and by an increased expression of VEGF-A and VEGF-C and VEGF receptors. Moreover, the average number of HPC expressing VEGFs was higher in samples with more extensive ductular reaction and angiogenesis. These findings are of interest because they are consistent with the idea that a VEGF-mediated cross talk between HPC/DR and endothelial cells may be involved in the remodeling of the vascular bed occurring in ductular reaction. The increased nutritional and functional demand is supported with changes in vascular architecture mediated by an increased secretion of VEGF. Furthermore, in PBC samples, reactive ductules were closer to fibrous septa and strands of ductular reactive cells penetrated in the cirrhotic nodules stimulating the formation of new vessels within fibrous septa. Interestingly, previous studies have shown that VEGF, released at the leading or lateral edge of developing fibrous septa, recruits activated hepatic stellate cells (HSC), which express VEGFR-1 and VEGFR-2 (21). In addition, *in vitro* experiments have shown that HSC migration was VEGFR-2-dependent through the activation of the Ras/ERK pathway. Furthermore, other studies have shown that *in vivo* administration of VEGFR-2 neutralizing antibody reduced neovascularization as well as fibrosis and the number of α-SMA positive cells in the chronic model of CCl_4-induced fibrosis (22).

Franchitto *et al.* did not find immunohistochemical evidence of VEGFR-2 expression in hepatic progenitor cells/oval cell. Unfortunately, the criteria used to distinguish HPC from reactive ductular cells remained unclear. Several studies in humans and rodent have shown that VEGFR-2 is expressed in reactive cholangiocytes. Strong expression of VEGFR-2 in cholangiocytes was reported in biliary atresia (23,24) , in ischemia/reperfusion damage (25) in chronic alcoholic liver disease (26), as well as in developing ductal plates (5). Furthermore, VEGFR-2 is expressed in cholangiocytes, in several animal models of cholangiopathies (17,18,22) both *in vitro* and *in vivo*, After administration of VEGF, VEGFR-2 is phosphorylated and induces cholangiocytes proliferation through the activation of the MEK/ERK1/2 pathway (12,13,19).

In conclusion, this study is consistent with the idea that VEGF is a major factor in liver repair through an autocrine effect on HPC/DR cell proliferation and a paracrine effect on surrounding endothelial cells. Exploring the complex interactions of HPC with surrounding inflammatory, mesenchymal and in the case of this study with endothelial cells will further enhance our understanding of physiologic and pathologic liver repair and, thereby, lead to new therapeutic possibilities.

Acknowledgements

Disclosure: The authors declare no conflict of interest.

References

1. Sell S. Liver stem cells. Science 1993;260:1224.
2. Crosby HA, Hubscher S, Fabris L, et al. Immunolocalization of putative human liver progenitor cells in livers from patients with end-stage primary biliary cirrhosis and sclerosing cholangitis using the monoclonal antibody OV-6. Am J Pathol 1998;152:771-9.
3. Strazzabosco M, Fabris L. Development of the bile ducts: essentials for the clinical hepatologist. J Hepatol 2012;56:1159-70.
4. Roskams TA, Theise ND, Balabaud C, et al. Nomenclature of the finer branches of the biliary tree: canals, ductules, and ductular reactions in human livers. Hepatology 2004;39:1739-45.
5. Fabris L, Cadamuro M, Libbrecht L, et al. Epithelial expression of angiogenic growth factors modulate arterial vasculogenesis in human liver development. Hepatology 2008;47:719-28.
6. Strazzabosco M, Fabris L, Spirli C. Pathophysiology of cholangiopathies. J Clin Gastroenterol 2005;39:S90-S102.
7. Coulomb-L'Hermin A, Amara A, Schiff C, et al. Stromal cell-derived factor 1 (SDF-1) and antenatal human B cell lymphopoiesis: expression of SDF-1 by mesothelial cells and biliary ductal plate epithelial cells. Proc Natl Acad Sci U S A 1999;96:8585-90.
8. Fabris L, Strazzabosco M, Crosby HA, et al. Characterization and isolation of ductular cells coexpressing neural cell adhesion molecule and Bcl-2 from primary cholangiopathies and ductal plate malformations.

Am J Pathol 2000;156:1599-612.
9. Fabris L, Cadamuro M, Guido M, et al. Analysis of liver repair mechanisms in Alagille syndrome and biliary atresia reveals a role for notch signaling. Am J Pathol 2007;171:641-53.
10. Loomes KM, Russo P, Ryan M, et al. Bile duct proliferation in liver-specific Jag1 conditional knockout mice: effects of gene dosage. Hepatology 2007;45:323-30.
11. Sparks EE, Perrien DS, Huppert KA, et al. Defects in hepatic Notch signaling result in disruption of the communicating intrahepatic bile duct network in mice. Dis Model Mech 2011;4:359-67.
12. Tan X, Yuan Y, Zeng G, et al. Beta-catenin deletion in hepatoblasts disrupts hepatic morphogenesis and survival during mouse development. Hepatology 2008;47:1667-79.
13. Fabris L, Cadamuro M, Fiorotto R, et al. Effects of angiogenic factor overexpression by human and rodent cholangiocytes in polycystic liver diseases. Hepatology 2006;43:1001-12.
14. Gaudio E, Barbaro B, Alvaro D, et al. Vascular endothelial growth factor stimulates rat cholangiocyte proliferation via an autocrine mechanism. Gastroenterology 2006;130:1270-82.
15. Simons M. An inside view: VEGF receptor trafficking and signaling. Physiology (Bethesda) 2012;27:213-22.
16. Spirli C, Locatelli L, Fiorotto R, et al. Altered store operated calcium entry increases cyclic 3',5'-adenosine monophosphate production and extracellular signal-regulated kinases 1 and 2 phosphorylation in polycystin-2-defective cholangiocytes. Hepatology 2012;55:856-68.
17. Spirli C, Okolicsanyi S, Fiorotto R, et al. Mammalian target of rapamycin regulates vascular endothelial growth factor-dependent liver cyst growth in polycystin-2-defective mice. Hepatology 2010;51:1778-88.
18. Spirli C, Okolicsanyi S, Fiorotto R, et al. ERK1/2-dependent vascular endothelial growth factor signaling sustains cyst growth in polycystin-2 defective mice. Gastroenterology 2010;138:360-371.e7.
19. Desmet VJ. Congenital diseases of intrahepatic bile ducts: variations on the theme "ductal plate malformation". Hepatology 1992;16:1069-83.
20. Franchitto A, Onori P, Renzi A, et al. Expression of vascular endothelial growth factors and their receptors by hepatic progenitor cells in human liver diseases. Hepatobiliary Surg Nutr 2012. [Epub ahead of print].
21. Novo E, Cannito S, Zamara E, et al. Proangiogenic cytokines as hypoxia-dependent factors stimulating migration of human hepatic stellate cells. Am J Pathol 2007;170:1942-53.
22. Yoshiji H, Kuriyama S, Yoshii J, et al. Vascular endothelial growth factor and receptor interaction is a prerequisite for murine hepatic fibrogenesis. Gut 2003;52:1347-54.
23. Strazzabosco M, Fabris L. Functional anatomy of normal bile ducts. Anat Rec (Hoboken) 2008;291:653-60.
24. Edom PT, Meurer L, da Silveira TR, et al. Immunolocalization of VEGF A and its receptors, VEGFR1 and VEGFR2, in the liver from patients with biliary atresia. Appl Immunohistochem Mol Morphol 2011;19:360-8.
25. Wang Z, Zhou J, Lin J, et al. RhGH attenuates ischemia injury of intrahepatic bile ducts relating to liver transplantation. J Surg Res 2011;171:300-10.
26. Morell CM, FL, Strazzabosco M. Vascular biology of the biliary epithelium. J Gastroenterol and Hepatology 2012. [Epub ahead of print].

Cite this article as: Spirli C, Strazzabosco M. Vascular endothelial growth factors in progenitor cells mediated liver repair. Hepatobiliary Surg Nutr 2013;2(2):65-67. doi: 10.3978/j.issn.2304-3881.2012.12.05

