

CASE REPORT

Rapidly increasing liver progenitor cell numbers in human regenerating liver after portal vein ligation and liver partition

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Abstract

Background: Liver regeneration is dependent on the proliferation of hepatocytes. Hepatic progenitor cells are intra-hepatic precursor cells capable of differentiating into hepatocytes or biliary cells. Although liver progenitor cell proliferation during the regenerative process has been observed in animal models of severe liver injury, it has never been observed *in vivo* in humans because it is unethical to take multiple biopsy specimens for the purpose of studying the proliferation of liver progenitor cells and the roles they play in liver regeneration. Associating liver partition and portal vein ligation for staged hepatectomy (ALPPS) is a staged procedure for inducing remnant liver hypertrophy so that major hepatectomy can be performed safely. This staged procedure allows for liver biopsy specimens to be taken before and after the liver begins to regenerate. **Case presentation:** The liver progenitor cell proliferation is observed in a patient undergoing ALPPS for a metastatic hepatic tumour. Liver biopsy is acquired before and after ALPPS for the calculation of average number of liver progenitor cell under high magnification examination by stain of immunomarkers. This is the first *in vivo* evidence of growing liver progenitor cells demonstrated in a regenerating human liver.

Keywords: liver regeneration, portal vein ligation, ALPPS

BACKGROUND

Human liver has a unique capacity to regenerate, mainly because of the ability of hepatocytes to change from a normal quiescent state to a growth state following liver trauma. Liver progenitor cells are precursors capable of differentiating into hepatocytes and biliary cells, and, therefore, play an important role in liver regeneration.¹⁻³ However, liver progenitor cell proliferation during the regenerative process has only been observed in animal models of severe liver injury because it is unethical to take multiple biopsy specimens from humans for the purpose of studying the proliferation of liver progenitor cells and the roles they play in liver regeneration.

Associating liver partition and portal vein ligation for staged hepatectomy (ALPPS) is a staged procedure for inducing remnant liver hypertrophy so that major hepatectomy can be performed safely.^{4,5} In the first stage, the right

portal vein is ligated and the right lobe of liver is divided to induce hypertrophy of remnant left lobe of liver. Then, after sufficient hypertrophy has occurred, major hepatectomy of the deceased liver is performed. This staged procedure allows for liver biopsy specimens to be taken before and after the liver begins to regenerate. We, therefore, took advantage of this approach to measure liver progenitor cell numbers before and during the regenerative process in a patient who required ALPPS for treatment of a metastatic liver tumour. Informed consent for participation in the study was obtained from participants.

CASE PRESENTATION

A 34-year-old male hepatitis B virus carrier with a history of recto-sigmoid colon cancer presented with metastatic liver tumours in both lobes. Small remnant liver volume (<35% of total liver volume) was predicted if major hepatectomy for bi-lobal

tumours was performed directly. Therefore, ALPPS was decided upon. One week after liver partition and portal vein ligation, liver volumetry revealed sufficient hypertrophic liver volume. Major hepatectomy was successfully performed at that time. A biopsy specimen of the left lobe was taken during the first stage and another was obtained during the secondary stage of ALPPS. The number of intrahepatic liver progenitor cells in the first-stage specimen was compared with that in the second-stage specimen. Liver progenitor cells usually appear in periportal and subcapsular areas of liver and can be identified by cytoplasmic staining with cytokeratin (CK)-7, CK-19 and neural cell adhesion molecule (N-CAM/CD-56). Therefore, sections were stained with cytokeratin immunomarkers and the number of liver progenitor cells in the periportal area was counted under high magnification. The average number of liver progenitor cells before portal vein ligation and liver partition was 11.4 cells per high-power field (x400) (Fig. 1). However, one week after the first stage of the ALPPS procedure, remnant liver volume had increased by 149.1% and the average number of liver progenitor cells had increased to 29.6

per high-power field (x400) (Fig. 1). To the best of our knowledge, this is the first evidence of rapidly growing liver progenitor cells in a regenerating human liver.

DISCUSSION

Liver progenitor cells have been shown to increase in number in regenerating livers in animal models of partial hepatectomy or drug-induced liver injury. However, it has never been directly observed in humans because it is unethical to take multiple biopsy specimens for the purpose of studying the proliferation of liver progenitor cells and the roles they play in human liver regeneration.

Portal vein embolization is a well-accepted method to induce liver regeneration under angiography. Major curative hepatectomy can then proceed four weeks after induced liver regeneration with sufficient remnant liver. ALPPS is another way to induce liver regeneration and allows for the measurement of changes in liver progenitor cell numbers in a regenerating liver. This is because liver biopsy specimens need to be taken during both stages of the procedure.

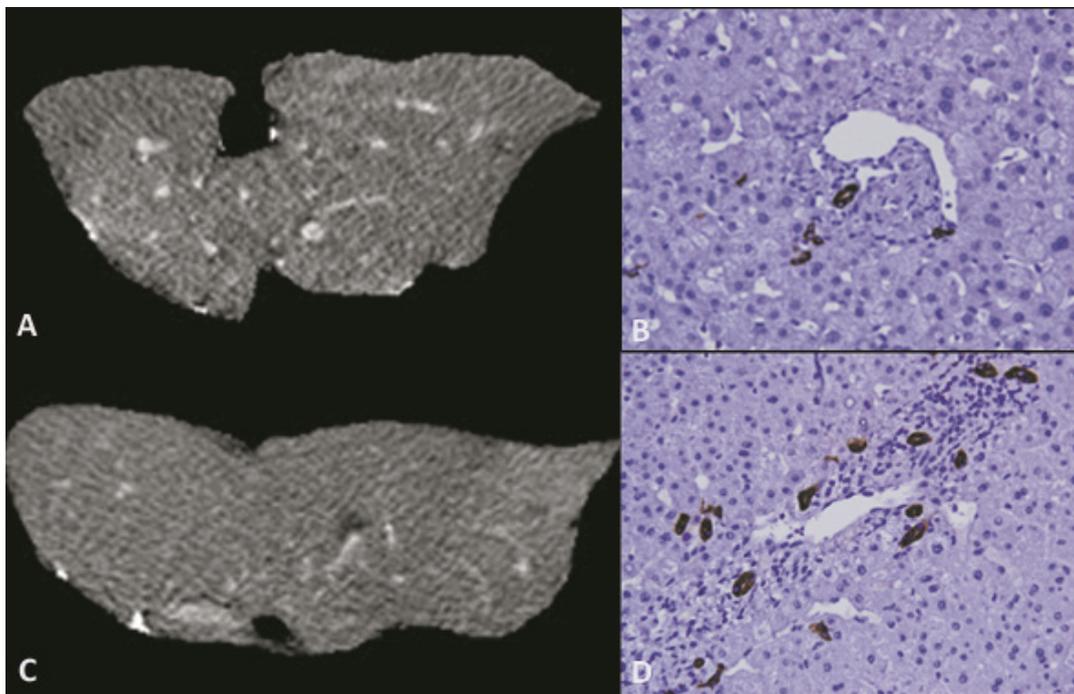


FIG. 1: (A) Volume of left lobe of liver before right portal vein ligation and liver partition, 355.2 cm³. (B) Liver progenitor cells in liver biopsy specimen of left lobe before right portal vein ligation and liver partition, average number of liver progenitor cells, 11.4/HPF (high-power field). (C) Volume of left lobe of liver 7 days after 1st stage of ALPPS, 529.6 cm³. (D) Liver progenitor cells in liver biopsy specimen of left lobe, 7 days after 1st stage of ALPPS, average number of liver progenitor cells, 29.6/HPF

Animal studies have shown that liver progenitor cells generate in the periportal region. Paku *et al* studied the origin and structural evolution of early proliferating liver progenitor cells in rats administered 2-acetylaminofluorene and found that liver progenitor cells were generated by proliferation of the terminal biliary ductules, which then formed ductular structures representing an extension of the canals of Hering.⁶ In the human liver, most liver progenitor cells are also generated in the periportal region. Liver progenitor cells are quiescent in normal liver and start proliferating in response to severe liver injury. Kym *et al* reported that liver progenitor cell numbers were directly related to the severity of chronic liver disease and that they were not observed in normal liver controls.⁷ In addition, Fausto *et al* reported that intra-hepatic precursor cells (liver progenitor cells) replicated rapidly when hepatocyte proliferation was blocked or delayed.² In our patient, a small number of liver progenitor cells was observed in the terminal portal region before portal vein ligation and liver partition; however, the number of cells in each region had increased by about 3 fold prior to performing the secondary stage of the ALPPS procedure.

Conclusion

To the best of our knowledge, this is the first study to provide *in vivo* evidence of rapidly growing liver progenitor cells in a regenerating human liver. However, more studies are needed to prove the relationship between liver progenitor cell proliferation and liver regeneration in humans.

ACKNOWLEDGEMENT

The authors would like to express sincere thanks to the coworkers of Changhua Christian Hospital, Transplant Medicine & Surgery Research Center.

Competing interests

The authors declare that they have no competing interests. No financial support is reported.

List of abbreviations used

ALPPS - associating liver partition and portal vein ligation for staged hepatectomy
 CK - cytokeratin
 N-CAM - neural cell adhesion molecule
 HPF - high-power field

Authors' contributions

The first two authors contributed equally to this work. K-H Lin carried out the conception and design of the study. H-T Hsu and T-H Teng analysed the pathology of liver tissue. Y-L Chen and P-Y Lin critically revised the manuscript and gave the final approval. All authors read and approved the final manuscript.

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