ADVANCES IN HEPATOLOGY

Current Developments in the Treatment of Hepatitis and Hepatobiliary Disease

Section Editor: Eugene R. Schiff, MD

Hepatocyte Transplantation

Ira J. Fox, MD
Director, Hepatocyte Transplant Program
Children's Hospital of Pittsburgh
Professor of Surgery
University of Pittsburgh School of Medicine
McGowan Institute for Regenerative Medicine
Pittsburgh, Pennsylvania

G&H What is the difference between hepatocyte transplantation and liver transplantation?

IF With liver transplantation, the entire organ is replaced. If there is acute or chronic rejection that compromises the liver and cannot be controlled, then the patient may require retransplantation. With hepatocyte transplantation, only a small fraction of liver cells—hepatocytes, the metabolic engine of the liver—are replaced, leaving intact all of the other types of cells within the liver, including endothelial cells, stellate cells, Kupffer cells, and fibroblasts. With this approach to transplantation, between 5% and 15% of the host's liver is replaced with transplanted hepatocytes.

G&H How is a hepatocyte transplantation procedure performed?

IF A catheter is placed into the main blood vessel that supplies nutrients to the liver. The catheter can be inserted through the skin over the liver into this portal vein, or an incision can be made around the belly button, where the remnant of the umbilical vein can be identified and reopened to gain access via catheter to the portal vein.

The hepatocytes that will be transplanted are isolated from livers that were donated for transplantation but were not used—perhaps because the ischemia time was too long, there was traumatic injury to the liver, or there were other concerns, such as fibrosis or too much fat in the liver. We isolate hepatocytes in a clean, US Food and Drug Administration–approved facility by using collagenase to digest the liver, a method similar to that used to separate islet cells from the pancreas for transplantation.

Once the cells are isolated, they are transplanted using a perfusion pump through the portal vein, and, through the spaces between endothelial cells, the hepatocytes enter the liver next to the native hepatocytes, where it is essentially impossible to distinguish the donor cells from the host cells.

G&H What are the indications for hepatocyte transplantation?

IF There are 2 major disease categories we now consider candidates for cell therapy. The first category includes patients with liver-based metabolic disorders that do not lead to cirrhosis. Examples include Crigler-Najjar syndrome, in which there is a defect in the enzyme that conjugates bilirubin; urea cycle disorders, in which there is abnormal processing of amino acids, leading to elevated ammonia levels; and phenylketonuria, in which phenylalanine is not processed properly. The second category includes patients with acute liver failure. In general, candidate conditions for hepatocyte transplantation are those in which the architecture of the liver is intact.

G&H Can liver failure from cirrhosis be treated with this approach?

IF Liver failure from cirrhosis, the most common indication for liver transplantation, cannot yet be treated by hepatocyte transplantation because the native liver is

structurally abnormal. Donor hepatocytes are not able to get into the liver because of a thickening of the extracellular matrix. Placing hepatocytes in extrahepatic locations, such as the spleen or in a lymph node, may be effective, but this approach has not been tested clinically. In the latest approach being investigated, a decellularized liver from an animal, such as a pig, is repopulated with viable hepatocytes and other types of liver cells, which can then be used as a bioartificial liver that could potentially be engrafted into the patient.

G&H What are the challenges with transplanting hepatocytes?

IF We do not have a good way of getting an adequate number of cells into the liver to correct most diseases. Isolated hepatocytes remain viable for approximately 48 hours after isolation and, therefore, must be transplanted within this time frame. However, transplanting all of the cells within 48 hours often exceeds the capacity of the portal vein to accept and disburse the cells into the liver. Portal hypertension may result, causing varices and possibly gastrointestinal bleeding. This, however, has never occurred. Thrombosis of the portal circulation could also result and could lead to liver dysfunction. Finally, new connections may theoretically be opened between the portal circulation and the systemic circulation, a process known as shunting, allowing hepatocytes to translocate to the lungs, which can cause cardiovascular instability or pulmonary insufficiency.

G&H What potential solutions to this problem are being investigated?

IF One possible solution is to use cells from multiple donors and to infuse them over a long period of time. There are also some diseases in which there might be a selective ability for donor hepatocytes to replace diseased host cells after transplantation. These diseases include alpha-1 antitrypsin deficiency, hereditary tyrosinemia, and Wilson disease. Unfortunately, by the time these patients are seen, they have already developed cirrhosis, so it is too late to treat them by hepatocyte transplantation.

However, in animal studies, low-dose irradiation to part of the liver has been shown to accomplish a similar result. First, radiation therapy causes short-term loss of the endothelial cells that line the portal vein, enabling the hepatocytes to more easily enter the liver. Second, it prevents the native hepatocytes from proliferating normally in response to injury or any other condition that would result in a regenerative signal. As a result, the donor hepatocytes can selectively grow when they are infused after the host liver is irradiated, allowing a greater extent of the host liver to be replaced by donor cells. This approach is being investigated in a clinical trial.

G&H What other challenges exist?

IF We have not yet seen long-term evidence of engraftment. The longest follow-up we have is a few years; we do not have longer-term follow-up data. With hepatocyte transplantation, it is difficult to identify transplanted cells by biopsy, which is routinely performed in organ transplant recipients, so it is difficult to know definitively whether the donor cells are being rejected until it is too late to intervene. This is also the case following islet transplantation. We are examining some experimental approaches for monitoring the status of the graft in order to predict rejection.

G&H Is it difficult to study hepatocyte transplantation in a clinical trial setting?

IF One major challenge is that the parents of patients with metabolic disorders view their children as stable and often are not interested in considering an experimental intervention, preferring instead to wait for an organ transplant. This appearance of stability in a patient with Crigler-Najjar syndrome or a urea cycle disorder is deceiving and at odds with the fact that a fatal episode of poor compensation could occur at any moment and lead to death or severe neurologic injury. Undergoing an experimental therapy could potentially provide enough enzyme activity to avoid such an outcome. Also, most families are hesitant to have their child be the first to undergo an experimental therapy, even though the process is reversible, has been performed in more than 100 patients worldwide, and has been associated with very few, if any, complications.

G&H How is the process reversible?

IF After hepatocytes are transplanted, if there is a significant infection or an immune suppression–related complication, immune suppression can simply be withdrawn, causing the cells to be rejected without symptoms and the patient's status to revert to how it was before the therapy was instituted. Also, if a patient decides to undergo a hepatocyte transplantation, his or her access to a donor organ and ability to receive an organ transplant is not affected.

G&H What happens to patients if hepatocyte transplantation is ineffective?

IF If a patient undergoes hepatocyte transplantation and the procedure does not adequately correct the liver abnormality, then liver transplantation is performed as originally planned. If a donated organ becomes avail-

able during the observation period following hepatocyte transplantation and the disease has not been corrected, we recommend that liver transplantation be performed.

G&H How has the experience been so far? Have hepatocyte transplantations been successful?

IF The experience worldwide has been similar from center to center. Crigler-Najjar syndrome and phenylketonuria have been corrected to the point where the bilirubin or phenylalanine levels, respectively, have been brought down to less than half of their initial values. We have not yet, however, seen cures for these conditions.

With Crigler-Najjar syndrome and urea cycle disorders, partial correction is not adequate because the patients remain at risk for severe brain injury. In almost all cases, the patients have ultimately received an organ transplant.

With phenylketonuria, partial correction of phenylalanine levels can result in very substantial improvement for patients. For most patients, the diet required to stay healthy is very restrictive. Lowering phenylalanine levels by more than half means that the diet can be less restrictive, and the risk of neurologic injury is significantly reduced. Thus, we believe that phenylketonuria is a good target disease for hepatocyte transplantation.

Dr Fox has no relevant conflicts of interest to disclose.

Suggested Reading

Dhawan A, Strom SC, Sokal E, Fox IJ. Human hepatocyte transplantation. *Methods Mol Biol.* 2010;640:525-534.

Dreyzin A, Barnato AE, Soltys KA, et al. Parent perspective on decisions to participate in a phase I hepatocyte transplant trial. *Pediatr Transplant.* 2014;18(1):112-119.

Gramignoli R, Tahan V, Dorko K, et al. New potential cell source for hepatocyte transplantation: discarded livers from metabolic disease liver transplants. *Stem Cell Res.* 2013;11(1):563-573.

Mazariegos G, Shneider B, Burton B, et al. Liver transplantation for pediatric metabolic disease. *Mol Genet Metab.* 2014;111(4):418-427.

Puppi J, Strom SC, Hughes RD, et al. Improving the techniques for human hepatocyte transplantation: report from a consensus meeting in London. *Cell Transplant.* 2012;21(1):1-10.

Soltys KA, Soto-Gutiérrez A, Nagaya M, et al. Barriers to the successful treatment of liver disease by hepatocyte transplantation. *J Hepatol.* 2010;53(4):769-774.

(continued from page 588)

Kahrilas PJ, McColl K, Fox M, et al. The acid pocket: a target for treatment in reflux disease? *Am J Gastroenterol.* 2013;108(7):1058-1064.

Kwiatek MA, Roman S, Fareeduddin A, Pandolfino JE, Kahrilas PJ. An alginate-antacid formulation (Gaviscon Double Action Liquid') can eliminate or displace the postprandial "acid pocket" in symptomatic GERD patients. *Aliment Pharmacol Ther.* 2011;34(1):59-66.

Rohof WO, Bennink RJ, Boeckxstaens GE. Proton pump inhibitors reduce the size and acidity of the acid pocket in the stomach. *Clin Gastroenterol Hepatol.* 2014;12(7):1101-1107.e1.

Rohof WO, Bennink RJ, de Ruigh AA, et al. Effect of azithromycin on acid reflux, hiatus hernia and proximal acid pocket in the postprandial period. *Gut.* 2012;61(12):1670-1677.

Rohof WO, Bennink RJ, Smout AJ, Thomas E, Boeckxstaens GE. An alginate-antacid formulation localizes to the acid pocket to reduce acid reflux in patients with gastro-esophageal reflux disease. *Clin Gastroenterol Hepatol.* 2013;11(12):1585-1591; quiz e90.

Thomas E, Wade A, Crawford G, Jenner B, Levinson N, Wilkinson J. Randomised clinical trial: relief of upper gastrointestinal symptoms by an acid pocket-targeting alginate-antacid (Gaviscon Double Action)—a double-blind, placebo-controlled, pilot study in gastro-oesophageal reflux disease. *Aliment Pharmacol Ther.* 2014;39(6):595-602.