Monitoring hepatitis C infection in the liver allograft

Bertus Eksteen MBChB PhD FRCP(London)

Chronic hepatitis C virus (HCV) infection-induced end-stage liver disease is the leading indication for liver transplantation and, in 2011, accounted for 1364 (23.5%) liver transplants performed in the United States. Treatment options for HCV are rapidly evolving, with realistic expectations of being able to cure the majority of patients in the very near future before the need for transplantation arises. Until such time, the status quo we are faced with is a large cohort of HCV cirrhosis patients who will require salvage with liver transplantation. The difficulty with hepatitis C post-transplantation is that reinfection of the allograft is virtually universal. Reinfection occurs with a wide range of clinical presentations ranging from the most severe form, fibrosing cholestatic hepatitis, which occurs very early after transplantation and invariably leads to early graft failure and a possible need for retransplantation or death, to a milder but still aggressive course in the majority of patients leading to bridging fibrosis and cirrhosis. The rate at which this develops is approximately 30% to 50% at five years without antiviral treatment (1). An essential element of managing post-transplant hepatitis C is to detect individuals who are at risk of progression at an early stage, defined by most studies as a Metavir score ≥ 2 , and commence antiviral treatment (1).

Our ability to detect at-risk patients and the early stages of liver fibrosis in the allograft has been contentious and fraught with difficulty. The reasons have been multifactorial. It is often difficult to distinguish in patients within the first year post-transplant whether abnormalities in liver biochemistry are due to recurrent HCV infection, acute cellular rejection, chronic allograft rejection or unrelated transplant complications such as cytomegalovirus infection, biliary strictures or vascular insufficiency of the allograft. This is further complicated by reports that, in the setting of normal liver biochemistry, significant liver pathology may be found in up to one-third of liver biopsies (2) – hence the current consensus guidelines to monitor HCV post-transplant with serial protocol liver biopsies (3). Although protocol liver biopsies are regarded as the gold standard, the exact timing of such liver biopsies are not clearly defined, with most centres opting for post-transplant biopsies at six months, 12 months and every year thereafter to monitor the allograft for HCV recurrence and the development of significant fibrosis to prompt antiviral treatment. Liver biopsies are invasive and carry a small but significant risk of major complications estimated to be approximately 0.5%. Liver biopsies are imperfect and are prone to sampling errors, and are dependent on the quality of the needle biopsy because biopsies of a sufficient length are required to make an accurate assessment of fibrosis.

Because of these limitations and the invasive nature of protocol biopsies, several groups have assessed other markers that might be used to judge the development of fibrosis within the allograft. In the current issue of the *Canadian Journal of Gastroenterology*, a retrospective study from the Toronto Liver Transplant Unit (Toronto, Ontario) by Tanaka et al (4) (pages 131-136) aimed to evaluate potential biomarkers that could be used as an alternative to protocol liver biopsies in patients with hepatitis C. The investigators assessed 242 consecutive patients who underwent liver transplantation in their centre for end-stage liver disease due to chronic HCV infection. Ninety-one patients were excluded from further analysis because of either poor

biopsy quality, patient or graft survival shorter than 12 months, biopsies that showed acute or chronic rejection, biliary complications or patients who were selected for early antiviral treatment within six months of transplant. They assessed patients for the development of significant fibrosis, defined by Metavir scoring ≥ 2 , in biopsies that were performed as part of their protocol biopsy regimen, which consists of biopsies at six, 12 and 24 months post-transplantation. Univariate analysis at six months post-transplant revealed that an elevated aspartate aminotransferase (AST) level, histological activity and stage 1 fibrosis on biopsy were associated with the development of significant progression to fibrosis at 12 months. In the multivariate analysis at six months, they found that a high AST level (for every 1 IU/L, OR 1.025; $P < 0.001$) and increasing donor age were independent risk factors for the progression to significant fibrosis at 12 months. They expanded their analysis to show that an AST cut-off of 82.0 IU/mL had a sensitivity of 88.2% and a specificity of 79.9% to detect patients who progressed to significant fibrosis. This was associated with a negative predictive value (NPV) of 98.2% and a positive predictive value of 35.7%. The predictive value of elevated AST levels only reached significance at six months and did not impact in the analysis at one or two years. Hence, Tanaka et al concluded that an elevated AST reading of 82.0 IU/mL may be used as cut-off to avoid a protocol biopsy at six months but that protocol biopsies at 12 months and 24 months proved essential in managing these patients.

The study by Tanaka et al (4) aimed to answer one of the most hotly debated questions in HCV management, which is how to predict the cause of the disease after transplantation. Given the apparent limitations of a liver biopsy, much work has been performed to improve its predictive ability by adding additional measures of liver dysfunction. Measurements of the hepatic venous-portal gradient (HVPG) in combination with liver biopsy have been shown to improve its predictive accuracy. An HVPG ≥ 6 mmHg at one year improved the ability to predict patients with severe HCV recurrence to 80%, compared with only 60% who were diagnosed on liver biopsy alone (5). Others have successfully used digital morphometry of collagen deposition as measured with Picrosirius red staining to improve standard histological assessment at levels comparable with HVPG measurements. Unfortunately, this technique is wholly dependent on adequate biopsy length and remains prone to sampling variation.

Several groups have sought to replace conventional invasive liver sampling with noninvasive assessments of allograft fibrosis. Assessment of direct serum markers of fibrosis, most notably the 3-M-ALG algorithm, successfully identified most patients with mild fibrosis at six months post-transplantation with an NPV of 79% and normal portal pressure with an NPV of 90%. Its predictive value at one year after transplantation had an area under the curve (AUC) of 0.9 to predict portal hypertension and an AUC of 0.78 to predict significant fibrosis, and proved superior to the AST/alanine aminotransferase ratio and AST platelet ratio (APRI) indexes (6). A histological predictor of fibrosis is the presence of interface hepatitis and is a biological explanation for the AST association detected by Tanaka et al (4). Increased AST levels have been incorporated into several algorithms to predict HCV progression. The Benloch index combined

Southern Alberta Transplant Clinic, Snyder Institute for Chronic Diseases, University of Calgary, Calgary, Alberta

Correspondence: Dr Bertus Eksteen, University of Calgary, 3280 Hospital Drive Northwest, Calgary, Alberta T2N 4N1.

Telephone 403-220-3719, fax 403-210-9657, e-mail b.eksteen@ucalgary

Received and accepted for publication February 14, 2013

prothrombin time, albumin:protein ratio, AST level and time from liver transplantation to predicted significant fibrosis (Metavir ≥ 2) with an AUC of 0.8. A cut-off of 0.2 yielded 95% accuracy for the exclusion of significant fibrosis (7). However, in a prospective cohort validation, the AUC fell to 0.68 AUC and NPV to 88%, respectively. Further scoring based on platelet count, international normalized ratio, AST level and time following liver transplantation showed a similar AUC (0.82) and was shown to be superior to the APRI score. Another strategy to gauge hepatic inflammation has been to measure serum levels of the chemokine CXCL10, which is secreted in the liver on injury and has been shown to correlate with HCV progression to significant liver fibrosis (8).

Noninvasive imaging techniques are increasingly being used in hepatology to detect the development of fibrosis. Both magnetic resonance imaging elastography and ultrasound transient elastography (Fibroscan, Echosens, France) have been used and correlate well with histological fibrosis stage and HVP. Ultrasound transient elastography is reported to detect significant fibrosis or portal hypertension (HVP ≥ 6 mmHg), with an AUC of 0.90 and 0.93, respectively. The authors (9) concluded that a cut-off value of 8.7 kPa had an NPV for significant fibrosis and portal hypertension of $>90\%$. Other studies have similarly reported encouraging results with imaging alone or in combination with either direct serum fibrosis markers or the AST/alanine aminotransferase ratio and APRI scores.

In summary, the data by Tanaka et al (4) are consistent with the growing need to find a suitable noninvasive replacement for protocol liver biopsies and highlights the importance of AST levels as an indicator of necroinflammation in the liver and the subsequent development of fibrosis. Noninvasive methods of fibrosis holds great promise; however, in many cases, these methods will need to be validated in prospective studies before we completely wave good-bye to the protocol liver biopsy.

REFERENCES

1. Berenguer M, Schuppan D. Progression of liver fibrosis in post-transplant hepatitis C: Mechanisms, assessment and treatment. *J Hepatol* 2012.
2. Abraham SC, Poterucha JJ, Rosen CB, Demetris AJ, Krasinskas AM. Histologic abnormalities are common in protocol liver allograft biopsies from patients with normal liver function tests. *Am J Surg Pathol* 2008;32:965-73.
3. Samuel D, Forns X, Berenguer M, et al. Report of the monothematic EASL conference on liver transplantation for viral hepatitis (Paris, France, January 12-14, 2006). *J Hepatol* 2006;45:127-43.
4. Tanaka T, Therapondos G, Selzner N, Renner EL, Lilly LB. Serum aspartate aminotransferase level and previous histopathological findings enable reduction of protocol liver biopsies after liver transplantation for hepatitis C. *Can J Gastroenterol* 2013;27:131-6.
5. Blasco A, Forns X, Carrion JA, et al. Hepatic venous pressure gradient identifies patients at risk of severe hepatitis C recurrence after liver transplantation. *Hepatology* 2006;43:492-9.
6. Carrion JA, Fernandez-Varo G, Bruguera M, et al. Serum fibrosis markers identify patients with mild and progressive hepatitis C recurrence after liver transplantation. *Gastroenterology* 2010;138:147-58.
7. Benlloch S, Berenguer M, Prieto M, Rayon JM, Aguilera V, Berenguer J. Prediction of fibrosis in HCV-infected liver transplant recipients with a simple noninvasive index. *Liver Transpl* 2005;11:456-62.
8. Berres ML, Trautwein C, Schmeding M, et al. Serum chemokine CXC ligand 10 (CXCL10) predicts fibrosis progression after liver transplantation for hepatitis C infection. *Hepatology* 2011;53:596-603.
9. Carrion JA, Navasa M, Bosch J, Bruguera M, Gilibert R, Forns X. Transient elastography for diagnosis of advanced fibrosis and portal hypertension in patients with hepatitis C recurrence after liver transplantation. *Liver Transpl* 2006;12:1791-8.