Late recurrent post-transplant primary biliary cirrhosis in British Columbia

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EM YOSHIDA, RA SINGH, RK VARTANIAN, DA OWEN, SR ERB, CH SCUDAMORE. Late recurrent post-transplant primary biliary cirrhosis in British Columbia. *Can J Gastroenterol* 1997;11(3):229-233. Late recurrent primary biliary cirrhosis (PBC) following orthotopic liver transplant remains a controversial topic. The first documented case of recurrence occurring in 16 patients transplanted for PBC and followed at the authors' institution for longer than one year is presented. A 54-year-old man transplanted for PBC developed a cholestatic pattern of enzyme elevation on post-transplant day (PTD) 1305. Repeat antimitochondrial antibody was strongly positive (1:300 to 1:400). A liver biopsy revealed severe bile duct damage, lymphocytic cholangitis, focal periductal noncaseating granuloma and minimal endotheliitis. Recurrent PBC was diagnosed. At the time of orthotopic liver transplant this patient received induction immunosuppression with OKT3 crossed over to cyclosporine (CsA), azathioprine (AZA) and prednisone. AZA was discontinued early and maintenance CsA tapered to a target trough level of 150 to 200 ng/mL by PTD 365. Prednisone was withdrawn by PTD 664. CsA levels during PTDs 1225 to 1305 (before elevation of hepatobiliary enzymes) were below target at 114 to 166 ng/mL. Of the 16 patients, all but three were maintained on CsA, AZA and prednisone. One was on CsA (trough levels on target) and AZA; the other two, including the patient with recurrent PBC, were on CsA only. The trough CsA level of the patient without recurrent PBC has been within the target range. The authors speculate that the underlying defect in immunoregulation in PBC persists post-transplant and that in the patient without recurrent PBC this defect was unmasked by lowered maintenance immunosuppression – allowing recurrence of PBC in a previously stable liver allograft.

Key Words: Primary biliary cirrhosis, Recurrent symptoms, Transplantation

Récurrence tardive d'une cirrhose biliaire primaire post-transplantation en Colombie-Britannique

RÉSUMÉ : La récurrence tardive d'une cirrhose biliaire primaire (CBP) après une transplantation hépatique orthotopique reste un sujet controversé. Le premier cas documenté de récurrence survenant dans un groupe de 16 patients transplantés pour une CBP, et suivis pendant plus d'un an à l'hôpital où exerce l'auteur, est présenté. Un Canadien français de 54 ans, transplanté pour une CBP à développé une cholestase caractérisée par une hausse des enzymes au jour 1305 post-transplantation. L'examen répété des anticorps antimitochondriaux était fortement positif (1:300 à 1:400). Une biopsie du foie a mis en évidence une grave atteinte des canaux biliaires, une cholangite lymphocytaire, un granulome focal péricanalaire non caséux et une endothéliite minimale. Un diagnostic de CBP récurrente a été posé. Au moment de la transplantation hépatique orthotopique, ce patient avait reçu un traitement immunosuppresseur initial avec de l'OKT3 croisé avec de la ciclosporine (CsA), de l'azathioprine (AZA) et de la prednisone. On a cessé d'administrer l'AZA tôt tandis que le traitement d'entretien à la CsA a été diminué progressivement jusqu'à un niveau cible minimal de 150 à 200 ng/mL au 365^e jour post-transplantation. On a supprimé la prednisone au 664^e jour post-transplantation. Les niveaux de CsA pendant la période allant du 1225^e jour au 1305^e jour post-transplantation (avant la hausse des enzymes hépatobiliaires) était inférieurs au point cible, soit entre 114 et 166 ng/mL. Des 16 patients, tous sauf trois ont continué à recevoir de la CsA, de l'AZA et de la prednisone. Un recevait de la CsA (niveau minimal adéquat) et de l'AZA ; les deux autres patients, y compris le patient souffrant d'une CBP récurrente, recevaient seulement de la CsA. Le niveau minimal de CsA du patient sans CBP récurrente se trouvait dans les limites fixées. Les auteurs ont émis l'hypothèse que l'anomalie sous-jacente dans l'immunorégulation, dans la CBP, persiste dans la période post-transplantation, et que dans le cas du patient sans CBP récurrente, cette anomalie avait été démasquée en diminuant le traitement d'entretien immunosuppresseur, permettant ainsi une récurrence de la CBP dans une allogreffe hépatique auparavant stable.

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Over the past 15 years orthotopic liver transplantation has proven to be efficacious in the management of patients with end-stage primary biliary cirrhosis (PBC) (1). Referral of these patients for transplantation assessment is now routine. Although it may be presumed by both patients and physicians that successful transplantation will completely cure the original disease, this point is controversial. In 1982 Neuberger et al (2) from the King's College Hospital Liver Unit (London, United Kingdom) reported three patients with cholestatic jaundice who were thought to have recurrent PBC. This initial report was followed up seven years later with a report of 13 patients, including the three originally reported, who were felt to have recurrent PBC (3). Nine of the 13 had liver biopsies that were interpreted as compatible with PBC recurrence. Other centres have since reported recurrent PBC (4-6). However, some centres have also reported a failure to find clear evidence of recurrent PBC in their patients (7,8). Furthermore, one of the patients reported by the King's College Hospital's group (3) was recently discovered to have chronic rejection – rather than recurrent PBC (9).

We report the first documented case of late recurrent PBC post-transplant at our centre. The discovery of this patient prompted a review of all our PBC patients who were transplanted more than one year ago in an attempt to discover any factors that may have contributed to post-transplant PBC recurrence.

METHODS

After identification of the patient reported below, records of the British Columbia Transplant Society were reviewed to identify all patients transplanted for PBC (including patients transplanted at other centres but followed in British Columbia). The Vancouver Hospital Transplant Clinic charts of all patients more than 365 days post-transplant as of February 1, 1996 were reviewed. According to the protocol of the transplant clinic, liver enzymes and whole blood trough cyclosporine (CsA) levels (monoclonal radioimmunoassay, Incstar Cyclotrac, Minnesota) were obtained every two weeks. The target trough CsA level for patients after post-transplant day (PTD) 365 is 150 to 200 ng/mL. Some patients had been converted from soft gelatin capsule CsA to the microemulsion formulation. According to Vancouver Hospital Transplant Clinic protocols, the target trough CsA level for patients 365 days after transplantation taking microemulsion formulation CsA is 120 to 150 ng/mL. Liver biopsies were obtained for persistent elevations in alkaline phosphatase (ALP), gamma-glutamyl transpeptidase (GGT), aspartate aminotransferase (AST) or alanine aminotransferase (ALT). All biopsies were examined for features of acute rejection, which have been previously described (10). Antimitochondrial antibody (AMA) was ordered at the discretion of the transplant physician. Immunosuppressive medication and liver enzymes were recorded from the time of last follow-up for all patient charts reviewed. As well, the last reported liver biopsy report for each patient was reviewed. Criteria for the diagnosis of recurrent PBC included

persistently elevated hepatobiliary enzymes, a positive AMA equivalent to a titre of 1:80 post-transplant and a liver biopsy with compatible histological features (11).

CASE PRESENTATION

A 54-year-old man of French-Canadian descent received a liver allograft for end-stage PBC 3.5 years previously. Before transplantation serum liver enzymes were ALP 232 U/L (normal 45 to 125 U/L); GGT 129 U/L (normal less than 50 U/L); total bilirubin 69 µmol/L (normal less than 22 µmol/L); AST 103 U/L (normal 19 to 38 U/L); and ALT 60 U/L (normal 10 to 55 U/L). Serology for AMA three years pretransplant was positive at a titre of 1:640; repeat AMA a year before transplant was positive at 1:320. Serology for antinuclear antibody and antismooth muscle antibodies were negative, as were viral markers for hepatitis B and C.

Orthotopic liver transplantation was undertaken with standard immunosuppressive induction consisting of OKT3 for nine days after transplantation, overlapped with CsA on PTD 7, prednisone 20 mg/day and azathioprine (AZA) 100 mg/day. Pathological examination of the explanted liver revealed micronodular cirrhosis, prominent bile ductular proliferation, periportal lymphocytic infiltrate, piecemeal necrosis and cholestasis, findings consistent with stage IV PBC. Early in the post-transplant period, AZA was discontinued secondary to leukopenia, and immunosuppression was continued with CsA and prednisone.

Maintenance immunosuppression was gradually tapered; by PTD 365, CsA was titrated to achieve a target whole blood trough level of 150 to 200 ng/mL, compared with 350 to 400 ng/mL in the first post-transplant month. Prednisone was slowly tapered from 20 mg/day and discontinued entirely by PTD 664 because of persistent obesity. Allograft function remained stable throughout PTD 1175 with no episodes of acute rejection and unremarkable liver enzymes at PTD 1175: ALP 97 U/L, GGT 69 U/L, AST 10 U/L and ALT 13 U/L. The CsA trough level from PTDs 1225 to 1305 was between 114 and 166 ng/mL. On PTD 1305 the hepatobiliary enzymes were noted to be elevated: ALP was 286 U/L, GGT 450 U/L, AST 63 U/L and ALT 63 U/L. Repeat liver chemistry on PTD 1309 revealed ALP to be 303 U/L, GGT 474 U/L and AST 77 U/L. An abdominal ultrasound did not reveal any dilated biliary ducts or stones, and a core liver biopsy was obtained on PTD 1311.

The liver biopsy (Figure 1) revealed minimal (and probably artefactual) evidence of endotheliitis, patchy portal-based mononuclear infiltrates and areas of severe bile duct damage with lymphocytic cholangitis accompanied by a focal portal-based periductal noncaseating granuloma. The biopsy was compatible with PBC based on histopathological features previously described (11). A repeat AMA was strongly positive at 3+ (equivalent to a titre of 1:300 to 1:400).

Because of possible acute rejection the patient was treated with pulse methylprednisolone 1 g intravenously daily for three days, followed by prednisone 20 mg/day. Follow-up serum liver enzymes gradually improved such that on PTD 1353, ALP was 118 U/L, GGT 267 U/L and AST 19 U/L.

Figure 1) **Left** Photomicrograph of the liver biopsy obtained on post-transplant day 1311. Accompanying the florid duct lesion (arrow) is a periductal noncaseating granuloma (hematoxylin and eosin, $\times 110$). **Right** A 5 μm step section that best demonstrates the portal-based noncaseating granuloma (arrow) (hematoxylin and eosin, $\times 110$)

Ursodeoxycholic acid was started at 500 mg tid. Serum liver chemistry on PTD 1402 revealed ALP of 88 U/L, GGT 115 U/L and AST 13 U/L.

On the basis of the acute cholestatic pattern of liver enzymes, strongly positive AMA and liver biopsy revealing changes consistent with PBC, but atypical for acute cellular rejection alone, a diagnosis of recurrent allograft PBC was made.

RESULTS

From 1989 until February 1, 1996, 23 patients transplanted for PBC (20 transplanted at the Vancouver Hospital, three transplanted in another province) were identified from the British Columbia Transplant Society records. Sixteen patients were identified who were PTD 365.

Maintenance immunosuppression for 13 patients consisted of CsA, AZA and prednisone. One patient was maintained on microemulsion formulation CsA (trough levels 141 to 223 ng/mL) and AZA, and two patients, including

the one described above, were maintained on CsA alone. The trough CsA level of the patient without recurrent PBC (on soft gelatin capsule CsA) was 153 to 221 ng/mL.

Nine of the 16 patients had normal liver enzymes (ALP, GGT, AST and ALT) throughout the last follow-up. Seven patients, including the patient reported with recurrent PBC, had elevated hepatobiliary enzymes. The six remaining patients with elevated hepatobiliary enzymes were all on triple immunosuppression. The first patient suffered from recurrent biliary strictures and portal vein thrombosis, and died secondary to complications of portal hypertension. The second patient with previously stable liver enzymes developed an acute elevation of hepatobiliary enzymes (ALP 287 U/L and GGT 362 U/L) on PTD 589. Liver biopsy revealed late acute rejection with no features of PBC. The third and fourth patients had chronically elevated hepatobiliary enzymes more than 1.5 times the upper limit of normal with negative follow-up AMA and previous documented acute rejection on liver biopsy during the first year. The third patient also

had a history of chronic post-transplant alcohol abuse. The fifth patient, post-transplant AMA status unknown, had persistently elevated hepatobiliary enzymes up to and including PTD 1931. The last liver biopsy at PTD 618 revealed acute rejection with significant fibrosis and decreased bile ducts. The sixth patient developed persistently elevated liver enzymes (ALP 228 U/L, GGT 365 U/L and AST 88 U/L) at PTD 1528. A liver biopsy was refused. She was started on ursodeoxycholic acid with complete normalization of liver enzymes by PTD 1703 (ALP 84 U/L, GGT 48 U/L, AST 28 U/L and ALT 28 U/L). A repeat AMA (ELISA) was positive at a titre of 1:320.

DISCUSSION

Although recurrence of original disease in liver allografts is well recognized in patients transplanted for viral hepatitis (12,13) and malignancy (14), it is less well appreciated for autoimmune diseases such as PBC. This is probably because allograft recipients are continued on potent immunosuppressive medications on a long term basis. Further, two of the drugs traditionally deemed the cornerstones of post-transplant immunosuppressive regimens, CsA and corticosteroids, have been demonstrated to be of efficacy to treat active PBC (15,16), albeit with significant drug-related adverse effects. The basic underlying defect in immunoregulation that produces PBC, however, is expected to persist after liver transplantation.

We diagnosed late recurrent PBC in our patient on the basis of elevated hepatobiliary enzymes, a strongly positive AMA at the time of diagnosis and the histological finding of a 'florid duct lesion', consisting of characteristic ductal epithelial changes with lymphocytic cholangitis accompanied by a periductal granuloma. The florid duct lesion on liver biopsy has been the characteristic finding in all series reporting recurrent post-transplant PBC (2-6, 17), although not all patients in these reports have had periductal granulomas (3,6). The AMA post-transplant classically falls to low titres in the early post-transplant period and may rise to higher titres as the post-transplant period progresses (3,7). Our patient with recurrent PBC had a strongly positive post-transplant AMA, which was similar to his pretransplant AMA. Although recurrent PBC has been described in allograft recipients with an undetectable AMA (5), we interpret the strongly positive AMA in our patient to support a diagnosis of recurrent PBC. In comparison, two of our patients with chronically elevated liver enzymes following episodes of rejection within the first transplant year had, on recent repeat testing, a negative AMA.

The possibility of allograft rejection must be considered when determining the diagnosis in a transplant recipient with inflammatory bile duct damage on biopsy. As occurs in PBC, biliary epithelium is also a prime target of rejection, which presents with biliary injury in its early stages (10). Indeed, a patient included in a widely cited study (3) of recurrent PBC was recently found, on examination of the previously transplanted explanted liver, to have chronic rejection rather than PBC (9). This patient did not have

portal granulomas in the original report (3). As well, Gouw and associates (8) could not find any histological differences in biliary damage between a group of patients transplanted for PBC and a non-PBC group on protocol biopsies. Despite these reports, and recognizing that there could be overlap in the histological features of rejection and recurrent PBC, there also appear to be features that allow distinction between the two conditions. These features are presence of endothelial inflammation (endotheliitis); and presence of a mixed portal infiltrate in rejection, versus the presence of the florid duct lesion with periductal granulomas and predominantly mononuclear portal-based infiltrates in recurrent PBC. Our patient clearly had a florid duct lesion (Figure 1) with a periductal granuloma which, in this clinical setting, is diagnostic of recurrent PBC. Granulomas in recurrent PBC are often periductal, with associated ductal damage, and occur many months to years after transplantation. Recently, Ferrell and associates (18) reported the presence of portal-based granulomas in two patients with acute cellular rejection. These two patients, however, were only a short time into the post-transplant period and the granulomas were located at the edge of the portal tract away from the bile duct. In their series, Ferrell et al had no difficulty in distinguishing acute rejection from recurrent PBC based on histological features.

Our patient is the first at our centre to develop documented late recurrent PBC, occurring 3.5 years post-transplant. Upon review of our patients transplanted for PBC, this patient was only one of two who had maintenance immunosuppression consisting solely of CsA. Furthermore, the CsA trough levels in the 2.5 months before the increase in hepatobiliary enzymes and confirmatory liver biopsy were below the therapeutic target. Because the primary immunological abnormality responsible for PBC can be expected to persist after transplantation, we speculate that reduced maintenance immunosuppression may have been a factor in the development of recurrent PBC. One of the current theories is that PBC is a disease of impaired immunoregulation (19). If an underlying defect in immunotolerance remains, then a reduction in immunosuppressive medication may unmask this defect by allowing activation of the immune system's effector cells. The role of maintenance immunosuppressive medications has not been adequately assessed in most studies of recurrent PBC. In the study of Gouw et al (8), in which no recurrent PBC was found in their transplant population, patients were maintained on CsA and a median prednisolone dosage of 17 mg/day at one year, 12 mg/day after two years and then 10 mg/day indefinitely. Most centres in North America would consider this regimen of corticosteroids to be high. On the other hand, prednisone was withdrawn in five of nine King's College Hospital patients with biopsy-proven recurrent PBC (2), and two centres reporting recurrent PBC withdrew maintenance corticosteroids after the first year (4) or sooner (6). Because prednisone has been effective in the treatment of mild PBC (16) we speculate that withdrawal of corticosteroids may be a factor in the appearance of recurrent PBC. Of interest, the King's College Hospital group has also

reported recurrent PBC in two patients maintained on FK506 (tacrolimus) (17) without corticosteroids.

CONCLUSIONS

We report the first case of recurrent PBC at our institution. The long term clinical significance of this disease recurrence is unknown. Because PBC is a chronic disease that progresses to end-stage liver disease over many years, the situation presumably is similar in the post-transplant patient. As in pretransplant PBC (20), ursodeoxycholic acid may be of some benefit in post-transplant recurrence.

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