Transpl Int (1994) 7 [Suppl 1]: S346–S349

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The impact of hepatitis C virus infection on liver disease in renal transplant recipients

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Abstract To assess the prevalence of hepatitis C virus (HCV) infection in renal transplant recipients and its impact on posttransplant liver disease, the sera from 176 recipients who had been followed for 1–20 years (mean 8.3 years) were tested for HCV-specific antibody using enzyme immunoassay. HCV-specific antibody was detected in 53 patients (30.1%) including 2 patients also positive for hepatitis B surface antigen (HBsAg). Among 167 HBsAg-negative patients, the presence of HCV-specific antibody was associated with an increased incidence of chemically significant hepatitis (70.6% vs. 9.5% in anti-HCV-negative patients, $P < 0.01$). Hepatitis was more likely to be chronic in anti-HCV-positive patients than in anti-HCV-negative patients ($P < 0.05$). Serious liver disease developed in 4 of 51 anti-HCV-positive, HBsAg-negative patients: liver failure causing death in 3 and hepatoma in 1. Liver biopsy specimens from anti-HCV-positive patients showed more aggressive histological lesions compared with those from anti-HCV-negative patients. We conclude that HCV infection is quite prevalent in our renal transplant recipients and plays a major role in posttransplant chronic liver disease.

Key words Renal transplantation
Hepatitis C virus infection
Chronic liver disease

Introduction

Chronic liver disease is a major problem in renal transplant recipients. It occurs in 6%–16% of patients, with considerable morbidity and mortality related to progressive liver damage [3, 15]. Of various potential etiological factors including viral infections and the immunosuppressive drugs themselves, non-A non-B hepatitis (NANBH) is believed to be the predominant cause of posttransplant chronic liver disease [4, 13].

In 1989, a blood-borne agent of NANBH, designated hepatitis C virus (HCV), was identified [6], and a recombinant-based immunoassay has been developed to detect antibodies to HCV (anti-HCV) [10]. Subsequently, HCV infection has been found to be the cause of the majority of NANBH transmitted by blood transfusion [2]. The purpose of this study was to determine the prevalence of anti-HCV in our renal ransplant recipients and the impact of HCV infection on posttransplant liver diesease.

Materials and methods

A total of 176 renal transplant recipients (95 men and 81 women) being followed during the period of the study (1989–1992) at the

Table 1 Demographic data of the patients

	Anti-HCV antibody		Significance
	(+)	(−)	
No. of patients	53 (30.1%)	123 (69.9%)	
Male/female	30/23	65/58	NS
Living-related donor/cadaveric donor	43/10	102/21	NS
HBsAg (+)	2 (3.8%)	7 (5.7%)	NS
Transfusion (+)	51 (96.2%)	111 (90.2%)	NS
Hemodialysis duration	44.0 months	25.1 months	$P < 0.01$
Follow-up	102.2 months	97.7 months	NS

Department of Urology, Hyogo Prefectural Nishinomiya Hospital, Japan, was included. All but two patients had been treated by hemodialysis before transplantation. Among them, 145 patients received living-related and 31 received cadaveric kidney allografts. While 162 patients had been transfused with at least two units of random blood before and during transplantation, 14 had never received a blood transfusion. The patients with a functioning graft have been followed up for 1–20 years after transplantation (mean 8.3 years). As maintenance immunosuppressive therapy, 73 patients received azathioprine and prednisolone, and 97 were given cyclosporine and prednisolone with or without a low dose of azathioprine. The remaining 6 patients were treated with FK 506, a new immunosuppressant, and prednisolone.

Serum samples were collected from the patients immediately before an assay and tested for anti-HCV using a first-generation enzyme immunoassay detecting antibody to the HCV C100-3 antigen (Ortho Diagnostic Systems) up to November, 1991, and thereafter by a second-generation enzyme immunoassay (Ortho Diagnostic Systems) which could detect antibodies to three recombinant antigens derived from the HCV genome. The vast majority of the patients as serially examined by both assay systems. Those who were anti-HCV-positive on at least one occasion were considered as positive.

Liver function tests, including alanine aminotransferase (ALT), aspartate aminotransferase (AST), bilirubin, and alkaline phosphatase, were routinely performed every month during the follow-up period. These tests were done at closer intervals when necessary. The hepatitis B surface antigen (HBsAg) status of the patients was determined prior to transplantation and whenever abnormal liver function was noted after transplantation. Nine patients were chronic carriers of HBsAg.

Chemically significant hepatitis was defined as an elevation of serum aminotransferase (ALT and/or AST) greater than 100 IU/l. Chronic hepatitis was defined as hepatitis persisting for more than 6 months.

Laparoscopy and liver biopsy were carried out in 26 patients who had chemically significant hepatitis. Eight patients underwent serial liver biopsies. Thus, a total of 39 liver specimens as obtained. Sections of formalin-fixed liver specimens were stained using standard histologic techniques and reviewed by the pathologist.

Statistical analysis was performed using the chi-square test or Student's *t*-test where appropriate. The differences were considered significant if $P < 0.05$.

Results

Prevalence of anti-HCV

Among 176 patients studied, anti-HCV was detected in 53 patient (30.1%), including 2 patients also positive for HBsAg. The demographic data of the anti-HCV-positive and anti-HCV-negative patients are shown in Table 1. The duration of hemodialysis before transplantation was significantly longer in the anti-HCV-positive patients than in the anti-HCV-negative patients ($P < 0.01$). There were no significant differences in the sex ratio, donor source, HBsAg status, proportion of transfused patients, or duration of follow-up posttransplant between the two groups.

Table 2 The incidence of hepatitis in relation to anti-HCV serology

	Anti-HCV antibody		Significance
	(+) $n = 51$	(−) $n = 116$	
Hepatitis	36 (70.6%)	11 (9.5%)	$P < 0.01$
Acute	4	5	
Chronic	32	6	

Liver disease

In order to focus on the impact of HCV infection on posttransplant liver disease, nine chronic carriers of HBsAg, seven of whom developed hepatitis (acute hepatitis in one and chronic hepatitis in six), were excluded from the following analyses on the possible correlation between the anti-HCV status and liver disease.

Among 167 HBsAg-negative patients, 47 developed chemically significant hepatitis. The prevalence of anti-HCV was 76.6% (36/47) in the patients with hepatitis as compared with 12.6% (15/120) in those without hepatitis ($P < 0.01$). Table 2 shows the incidence of hepatitis in relation to anti-HCV serology. The presence of anti-HCV was associated with an increased incidence of hepatitis (70.6% in the anti-HCV-positive patients vs. 9.5% in the anti-HCV-negative patients; $P < 0.01$). Among those who had hepatitis, chronic hepatitis was documented in 32 of 36 (88.9%) anti-HCV-positive patients compared with 6 of 11 (54.5%) anti-HCV-negative patients ($P < 0.05$). Serious liver disease developed in 4 of 51 anti-

Table 3 Serious liver disease in anti-HCV-positive patients (*AH* acute hepatitis, *CH* chronic hepatitis)

Case	Liver disease (period from transplant to onset; months)	Prognosis
LD 44	CH (1.5) → Hepatoma (168)	Alive (181)
LD 113	AH (6) → Liver failure	Dead (11)
LD 159	CH (14) → Liver failure	Dead (53)
LD 169	CH (2) → Liver failure	Dead (57)

Table 4 Histological examination results

Diagnosis	Anti-HCV antibody	
	(+) $n = 28$ [a]	(−) $n = 11$ [b]
Normal liver	3	1
Fatty infiltration	3	1
Drug-induced hepatitis	5	5
Reactive hepatitis	3	2
Chronic persistent hepatitis	10	1
Chronic active hepatitis	4	1
Hemosiderosis	2	–

[a] Biopsies taken from 19 patients
[b] Biopsies taken from 7 patients

HCV-positive patients: hepatoma in 1 and liver failure causing death in 3 (Table 3).

The incidence of hepatitis was higher in the patients treated with azathioprine (AZ) than in those treated with cyclosporin A or FK 506 (CsA/FK) irrespective of anti-HCV serology (AZA vs. CsA/FK; 79.2% vs. 63.0% in the anti-HCV-positive, and 14.6% vs. 6.7% in the anti-HCV-negative patients). However, the differences were not statistically significant.

Liver histology

Of the 39 liver biopsy specimens available, 28 were obtained from 19 anti-HCV-positive patients, and 11 were from 7 anti-HCV-positive patients. Histological examination revealed that the anti-HCV-positive patients had more aggressive liver disease than the anti-HCV-negative patients (Table 4). In the anti-HCV-positive group, chronic persistent hepatitis (CPH) and chronic active hepatitis (CAH) were confirmed in 10 (35.7%) and 4 (14.3%) of 28 specimens, respectively, whereas drug-induced hepatitis was the predominant histological lesion in the anti-HCV-negative group, and only 2 of 11 (18.2%) specimens showed CPH and CAH. There were no specimens showing histological evidence of cirrhosis in either the anti-HCV-positive or anti-HCV-negative group. In the 8 patients with serial liver biopsies, 3 of 5 (60.0%) anti-HCV-positive patients and 1 of 3 (33.3%) anti-HCV-negative patients showed histological progression. The former 3 with a histologically normal liver or drug-induced hepatitis on initial biopsy subsequently developed CPH or CAH, while the liver of the latter patient changed from normal to reactive hepatitis between the initial and final biopsy.

Discussion

The prevalence of anti-HCV in our patients was 30.1%, much higher than that for blood donors in Japan (1.3%). The reported prevalence of anti-HCV in renal transplant recipients has ranged from 6.2% to 65.8% [1, 5, 9, 11, 12]. The differences may depend on the incidence of HCV infection in the general population, the profile of the patient population studied, or the accuracy of the assay used. It has also been reported that anti-HCV seroconversion occurred during the posttransplant follow-up period and that anti-HC positivity decreased with time after transplantation [9, 12]. Therefore, the prevalence of anti-HCV may be relatively low when serum samples currently collected are used for an assay compared wit the frozen-stored sera collected at the time of transplantation.

The prevalence of anti-HCV in the patients who developed hepatitis was 76.6% in contrast to 12.5% in those without hepatitis ($P < 0.01$). The incidence of hepatitis was 70.6% in the anti-HCV-positive patients and 9.5% in the anti-HCV-negative patients ($P < 0.01$). Anti-HCV-positive hepatitis was more likely to be chronic than anti-HCV-negative hepatitis ($P < 0.05$). These results clearly demonstrate a significant correlation between the anti-HCV status and posttransplant chronic liver disease. Since the presence of anti-HCV is considered a marker of ongoing rather than past infection [2, 8, 14], HCV is a major factor causing liver disease in renal transplant patients.

Furthermore, histological findings confirmed the impact of HCV infection on posttransplant liver disease. Anti-HCV-positive hepatitis patients had more aggressive histological forms of liver disease compared with anti-HCV-negative hepatitis patients. Histological progression was more frequently observed in anti-HCV-positive patients. Interestingly, no liver specimens showed cirrhosis in either group. This could be attributed to the administration of prednisolone, which is known to prevent fibrosis. Although the incidence of hepatitis was not correlated with the immunosuppressive regimen, it remains to be determined whether immunosuppressive

drugs including AZA CsA and FK506, which are hepatotoxic themselves, complicate the clinical and histological features of HCV infection.

Concerning the clinical outcome of HCV infection in renal transplant recipients, Huang et al. [9] reported that none of their HBsAg-negative, anti-HCV-positive patients developed either liver cirrhosis or liver failure during the 50.4 months of follow-up and suggested that HCV infection alone had a more benign clinical outcome compared with HBsAg-positive, anti-HCV-positive patients. In contrast, 4 of our 51 HBsAg-negative, anti-HCV-positive patients developed serious liver disease. Oliveras et al. [11] found that all advanced cases of liver disease in their anti-HCV-positive patients developed 5 years or more after transplantation. Long-term follow-up is needed to define the final prognosis of HCV infection.

It has been demonstrated that interferon is useful in the treatment of patients with chronic hepatitis C [7]. However, it is unknown whether interferon therapy is effective for hepatitis C in such immunosuppressed hosts as renal transplant patients and whether the therapy induces acute graft rejection. To assess these questions, we are now conducting a clinical trial of recombinant interferon alpha in a selected cohort of our renal transplant patients with chronic hepatitis C.

In conclusion, HCV infection is quite prevalent in our renal transplant recipients and plays a major role in posttransplant chronic liver disease. Further study is required to understand and to control HCV-related chronic liver disease better.

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