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J. Nery · V. Esquenazi · D. Weppler
C. Gomez · R. Cirocco
H. Gharagozloo · K. Zucker
R. Reddy · J. Casella · I. Parker
W. Faria · L. Jeffers · M. Carreno
J. Smith · M. Markow · M. Allouch
S. Babischkin · G. Bourke · M. Hill
E. Schiff · S. LaRue · J. Miller

# Hepatitis C in liver transplantation: preliminary study of prognostic factors

J. Nery (✉) · D. Weppler · R. Cirocco
K. Zucker · J. Casella · W. Faria
M. Carreno · M. Markow · S. Babischkin
V. Esquenazi · C. Gomez · H. Gharagozoo
J. Smith · M. Allouch · G. Burke · J. Miller
Division of Transplantation,
Department of Surgery,
University of Miami School of Medicine,
Miami, FL 33136, USA

R. Reddy · T. Parker · L. Jeffers · M. Hill
S. LaRue · E. Schiff
Center for Liver Diseases,
University of Miami School of Medicine,
Miami, FL 33136, USA

**Abstract** At the University of Miami liver transplantation for chronic liver disease in HCV-positive patients has shown good results, with a 92% patients survival rate (follow up 8 to 57 months, median 21). None the less, we found that a large number of patients are expected to develop serious histological graft damage and may need retransplantation, which may place a further strain on the already scarce donor resources.

We have conducted a preliminary investigation on the importance of parameters which may correlate with the prognosis of HCV grafts. We found no impact of HLA match or typing. An interesting hypothesis, which deserves further investigation, is that some HCV strains could be more virulent than others and play a role as an independent risk factor. We have identified six strains among our patients and the BK serotype shows a trend to be associated with a worse outcome. We have found that patients developing and maintaining higher liver enzyme levels (ALT and GGT) after transplant and those with higher levels of viremia may be at risk to develop serious damage to their grafts.

**Key words** Hepatitis C · Liver transplantation

## Introduction

End-stage liver disease related to hepatitis C virus (HCV) has become one of the most frequent indications for liver transplant (LTx). It is apparent that HCV recurs in many patients after LTx and that others may acquire the virus perioperatively [1, 2, 5–7, 10]. However, factors predicting the severity and outcome of the graft infection remain to be determined. This study was undertaken in order to understand the course and outcome of HCV infection in patients receiving LTx, and to investigate markers and risk factors for development of HCV-related disease in the liver graft.

## Patients and methods

Anti-HCV antibody and HCV RNA were investigated in samples of serum, frozen at −20 °C, obtained prior to transplant and postoperatively from 94 patients undergoing 110 LTx at the University of Miami between March 1987 and January 1993. A recombinant immunoblot assay (RIBA II) [8] was used for anti-HCV testing, and a qualitative reverse transcriptase polymerase chain reaction (rt-PCR) [9] was used for HCV RNA determination. Patients were considered to be HCV positive if one or both of these tests were positive. All the patients had serial liver biopsies and HCV-positive patients were classified in to three groups according to the most prevalent histological alterations. This classification excluded the features compatible with rejection, and three groups were defined: I (no histological changes, ballooning degeneration, mild to moderate steatosis and/or acidophilic necrosis); II (severe steatosis and/or acidophilic necrosis, mild to moderate focal acute hepatitis, chronic

persistent hepatitis); and III (severe acute hepatitis, piecemeal necrosis, chronic active hepatitis, bridging fibrosis).

Serum levels of liver enzymes, alanine aminotransferase (ALT) and gamma glutamyl transferase (GGT), and levels of viraemia were investigated as hypothetical markers for HCV disease. Correlations of mean ± SD values of ALT and GGT were longitudinally determined within the three groups and compared with a control group of 13 HCV-negative patients who also received LTx for chronic end-stage liver disease and who were similar to the HCV-infected patients with regard to age, sex, race and UNOS status. A total of 96 serial postoperative serum samples from 19 patients with detectable HCV RNA were analysed with a quantitative rt-PCR [4] that determined the number of viral copies (million/ml) in each sample. In addition, HCV strains were investigated in 11 HCV RNA-positive patients by DNA sequence variation in the relatively conserved 5′ untranslated region of the HCV RNA by rt-PCR followed by restriction fragment length polymorphism [11]. Finally, HLA typing and compatibility were analysed. The data were statistically evaluated by analysis of variance and Duncan's multiple range test.

## Results

HCV infection was found in 25 patients (27%) (15 males and 10 females; 23 Caucasians and 2 blacks) ranging in age from 9 to 65 years (median 47 years), of whom two acquired the virus peri- or postoperatively. End-stage chronic liver disease due to postnecrotic cirrhosis was the indication for LTx in all cases, and the following associated conditions were observed: prolonged alcohol abuse (four), hepatocellular carcinoma (two), hepatitis B (one), cystic fibrosis (one), $\alpha_1$-antitrypsin deficiency (one) and sclerosing cholangitis (one). Two patients died 1 and 17 months post-LTx from *Candida* sepsis and pneumonia, respectively, and 23 patients (92%) were alive and out of hospital with follow-ups ranging from 8 to 57 months (median 21 months). Three patients had to be retransplanted, two due to primary non-functioning grafts (day 3 and 16) and one due to recurrent hepatitis C resulting in severe graft dysfunction 3 months postoperatively.

Serologically, HCV recurred early after LTx (1–8 weeks) in 22 patients. Two acquired de novo infection and seroconverted at 1 and 17 months postoperatively. One patient who preoperatively was RIBA II and PCR positive remained negative 22 months after transplant. Another patient, also positive by both methods before LTx had become PCR negative at 16 months post-transplant, but his RIBA II remained positive.

The patient who died 1 month after transplant was excluded from further analysis. Histologically, seven patients (29%) were classified in group I, seven (29%) in group II and ten (42%) in group III. Patient follow-up in groups I, II and III were 24±14, 29±14 and 20±9 months, respectively, and the differences were not statistically significant.

ALT and GGT values for the control and the three groups are displayed in Fig. 1. ALT levels in group III were significantly higher than in the other groups, and GGT levels also correlated with severity of histological abnormalities. There was a tendency for patients in group III to have a higher number of viral copies than patients in groups I and II. However, the differences were not significant owing to the small sample size (Table 1). Six different HCV genotypes were identified among 11 HCV RNA-positive patients in three groups, as shown in Table 2. It was not possible in this study to establish a correlation between any individual strain and severity of infection. The number of HLA matches was not significantly different among the three groups nor was there any correlation with a specific A, B or DR type.

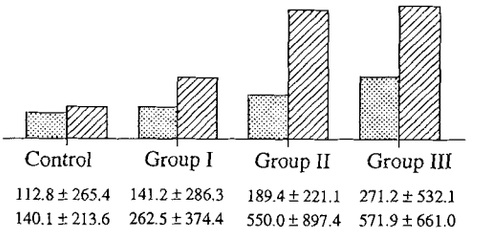


**Fig. 1** ALT and GGT values for the control group (C) and three patient groups (▨ ALT, C and I < II < III, $P < 0.05$; ▧ GGT, C < I < II and III, $P < 0.05$)

**Table 2** HCV strains

| Group | BK | HUTCH | HCV-J | J8 | JK1 | MIAMI |
|---|---|---|---|---|---|---|
| I | 1 | 1 | 0 | 0 | 0 | 0 |
| II | 1 | 0 | 1 | 0 | 1 | 0 |
| III | 3 | 1 | 0 | 1 | 0 | 1 |

**Table 1** Number of viral copies (million/ml)

| Group | n[a] | < 5 | 5 to < 10 | 10 to 50 | 50 to < 100 | 100 to 500 |
|---|---|---|---|---|---|---|
| I (6 patients) | 31 | 23 | 1 | 6 | 0 | 1 |
| II (3 patients) | 23 | 13 | 4 | 1 | 3 | 2 |
| III (10 patients) | 42 | 20 | 6 | 12 | 1 | 3 |

[a] Number of serum samples studied

## Discussion

The incidence of chronic liver disease resulting from HCV infection is high, being estimated at 25% or more [3], but the disease is believed to progress at a slow rate and development of cirrhosis may take many years. HCV recurrence is frequent after LTx and de novo infection has also been recognized. These facts are very important because the disease may progress faster in an immunosuppressed host. Despite this unfavourable aspect, the incidence of early graft failure was very low and both survival rate and quality of life were currently very good in these patients, which confirms that HCV-related chronic liver disease is a good indication for LTx. In this study, we observed that HCV recurrence was almost universal after LTx. On the other hand, a significant number of patients developed histological changes compatible with viral attack of their transplants which may progress to end-stage chronic liver disease, and ultimately result in graft loss.

The identification of indicators and risk factors for development of serious HCV-related disease in the liver allograft could allow the development of improved strategies to prevent graft dysfunction and loss. In this study, we found that patients who ultimately developed serious liver damage usually had higher levels of ALT and GGT. Also, there was apparently a correlation between high levels of viraemia and severity of histological changes, but a study analysing larger numbers of samples per patient is necessary to confirm this finding.

The preliminary analysis of different HCV genotypes showed an interesting tendency towards an association of the BK strain with an unfavourable prognosis, but the data are not conclusive owing to the small number of cases; however, this issue certainly deserves further investigation. The possibility that degrees of virulence in the genotypic varieties of HCV as an independent or contributory risk factor certainly deserves further investigation. Finally, we could not find any correlation between tissue typing or a specific HLA genotype and outcome in this group of patients.

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