American Journal of Transplantation 2013; 13: 1601–1605
Wiley Periodicals Inc.

doi: 10.1002/ajt.12209

Case Report

Sofosbuvir and Daclatasvir Combination Therapy in a Liver Transplant Recipient With Severe Recurrent Cholestatic Hepatitis C†

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Recurrent HCV infection following liver transplantation can lead to accelerated allograft injury that is difficult to treat with interferon. The aim of this study is to describe the first ever use of an interferon-free, all oral regimen in a liver transplant recipient with severe recurrent HCV. A 54-year-old male with HCV genotype 1b developed severe cholestatic HCV at 6 months posttransplant with ascites, AST 503 IU/mL, alkaline phosphatase of 298 IU/mL, HCV RNA of 12 000 000 IU/mL, and histological cholestasis with pericellular fibrosis. Sofosbuvir, an HCV polymerase inhibitor (400 mg/day), and daclatasvir, an HCV NS5A replication complex inhibitor (60 mg/day), were co-administered for 24 weeks. Within 4 weeks of initiating treatment, serum HCV RNA levels became undetectable and liver biochemistries normalized with concomitant resolution of ascites. The patient achieved a sustained virological response with undetectable HCV RNA at 9 months posttreatment. During and following treatment, the daily dose and blood level of tacrolimus remained stable and unchanged. The rapid and sustained suppression of HCV replication in this liver transplant recipient provides great promise for the use of combination oral antiviral regimens in other immunosuppressed and interferon refractory HCV patients.

Key words: Calcineurin inhibitors, direct-acting antiviral agents, hepatitis C, polymerase inhibitors

Abbreviations: AST, aspartate aminotransferase; ALT, alanine aminotransferase; DCV, daclatasvir; HCV, hepatitis C virus; LT, liver transplantation; WBC, white blood count.

Received 30 November 2012, revised 10 January 2013 and accepted 31 January 2013

Introduction

Hepatitis C virus (HCV) infection is the leading indication for liver transplantation (LT) in the United States and many parts of the world (1). However, patient survival is significantly lower in HCV patients compared to other recipients due, in part, to the inevitable recurrence of HCV infection in the allograft that is characterized by high levels of HCV replication that can lead to accelerated necroinflammation and fibrosis (2,3). Peginterferon and ribavirin combination therapy has been used in selected LT recipients with moderate to severe recurrent HCV but is limited by frequent side effects and low antiviral efficacy (4,5). Although the protease inhibitors, boceprevir and telaprevir, markedly improve response rates when combined with peginterferon and ribavirin in HCV genotype 1 patients, both of these drugs are substrates and inhibitors of CYP3A4 which can lead to potentially severe drug–drug interactions when co-administered with calcineurin inhibitors (6,7). Preliminary data indicate that these drugs are associated with a high rate of anemia, infection and poor tolerability in LT recipients (8,9). As a result, use of these agents with peginterferon and ribavirin is currently not recommended in LT recipients until further studies have been completed. Daclatasvir (DCV) is a potent NS5A replication complex inhibitor with demonstrated antiviral activity in HCV genotype 1 patients when co-administered with peginterferon and ribavirin (10,11). Similarly, sofosbuvir (GS-7977), a potent oral nucleotide analogue inhibitor of HCV polymerase activity, has shown great promise when combined with ribavirin with or without peginterferon in both naïve and previously treated patients with multiple HCV genotypes (12,13). Both of these investigational agents are orally dosed once a day and have had a generally favorable side effect profile. In addition, neither drug is anticipated to cause significant drug–drug interactions when co-administered with other CYP3A4 substrates (14,15). The aim of this study is to report the first ever use of sofosbuvir combined with daclatasvir in a

LT recipient with severe recurrent cholestatic HCV infection.

Results

A 54-year-old African American male with diabetes and coronary artery disease developed liver failure due to severe HCV genotype 1b infection that was resistant to prior interferon monotherapy and peginterferon and ribavirin combination therapy given pretransplant. At the time of LT, he had refractory ascites with a laboratory MELD score of 24. Within 6 months of LT, he developed severe recurrent cholestatic HCV infection with an HCV RNA level of 12 000 000 IU/mL, a serum aspartate aminotransferase (AST) of 503 IU/L, alanine aminotransferase (ALT) of 584 IU/L, alkaline phosphatase of 298 IU/L, total bilirubin of 1.9 mg/dL (direct 1.5 mg/dL) and INR of 1.3. He was fatigued with moderate ascites on exam and his body mass index was 22.2 kg/m^2. An ERCP at month 4 postLT showed no evidence of a biliary stricture, a liver ultrasound with Doppler excluded portal vein thrombosis, and an inferior venacavagram with pressure measurements at month 6 postLT showed no evidence of venous outflow obstruction. A liver biopsy demonstrated mildly active chronic hepatitis C with focal bridging fibrosis and diffuse swelling of hepatocytes with perisinusoidal collagen deposition, both common features of fibrosing cholestatic hepatitis (16) (Figure 1).

Despite early discontinuation of mycophenolate mofetil and minimizing his tacrolimus and prednisone, his clinical status continued to deteriorate. Following the approval of an emergency IND by the FDA (#113 500), the patient signed a written informed consent approved by the local IRB to receive DCV, 60 mg/day (Bristol-Meyers Squibb, Princeton, NJ) and sofosbuvir 400 mg/day (Gilead Sciences, Foster City, CA) for 24 weeks at the University of Michigan Institute for Clinical and Health research Unit (Ann Arbor, MI). The research protocol included a pretreatment baseline visit as well as planned study visits at weeks 1, 2, 3, 4 and every 2 weeks through week 24 as well as at posttreatment weeks 4 and 24 wherein routine labs and tacrolimus levels were obtained as well as monitoring for adverse events. Study medications were dispensed on a monthly basis and the patient kept a log of daily medication administration as well as all concomitant medications. Serum HCV RNA was measured using the Roche Cobas Taqman assay (lower limit of quantification = 43 IU/mL) at weeks 0, 1, 2, 4, 12 and 24 and months 1, 2 and 6 posttreatment.

On the first day of drug dosing, his serum creatinine was 1.0 mg/dL, hemoglobin 13.9 g/dL, white blood cell count (WBC) 4.4 $\times$ 10^3/mL and platelets were 166 $\times$ 10^3/mL. Concomitant medications included furosemide, pantoprazole, vitamin D, tacrolimus 1.5 mg twice a day, insulin NPH 32 units each AM and prednisone 3 mg/day. The patient was IL28-B genotype CT. Within 4 weeks of starting treatment, HCV RNA was below the lower limit of detection (i.e. $<$43 IU/mL) and his liver biochemistries normalized (AST 24, ALT 17, alkaline phosphatase 96, total bilirubin 0.8) along with improvement of his albumin to 4.3 g/dL and INR of 1.0 (Figure 2). In addition, his symptoms of fatigue and ascites resolved.

The patient completed 24 weeks of sofosbuvir and daclatasvir combination therapy with persistently undetectable HCV RNA and normal liver biochemistries. The patient did not experience any adverse laboratory or clinical adverse events attributed to either study drug. In fact with improvement in his fatigue, he was able to return to work by month 3 of antiviral treatment. At the end of treatment, his hemoglobin was 15.0 g/dL, WBC 7.7 $\times$ 10^3/mL, platelets 202 $\times$ 10^3/mL, INR 1.0 and creatinine was 1.3 mg/dL. The tacrolimus dose remained unchanged throughout antiviral treatment with a stable serum trough

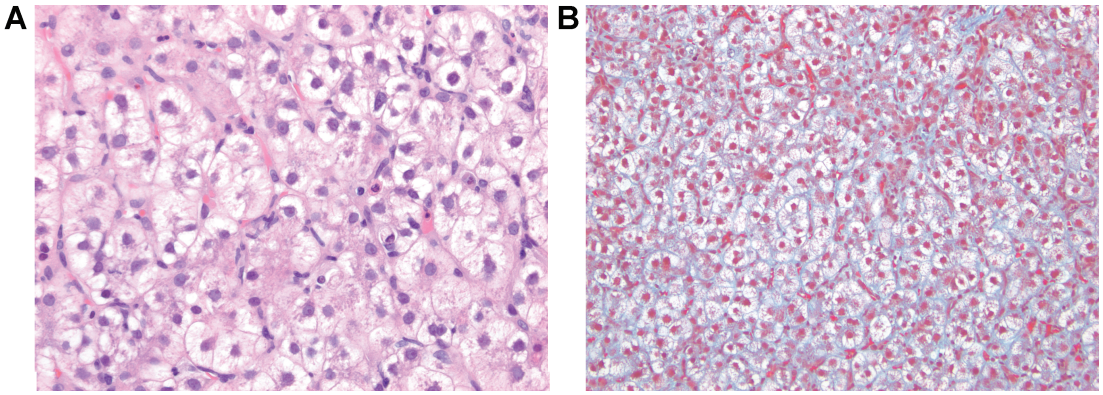

Figure 1: A: The hepatocytes are swollen with pale cytoplasm and arranged in clusters rather than cords, a common finding in fibrosing cholestatic HCV. The only evidence of inflammation is the few sinusoidal lymphocytes (hematoxylin and eosin 400$\times$). B: Fine blue collagen fibers separate the clusters of pale swollen hepatocytes (trichrome stain, 200$\times$).

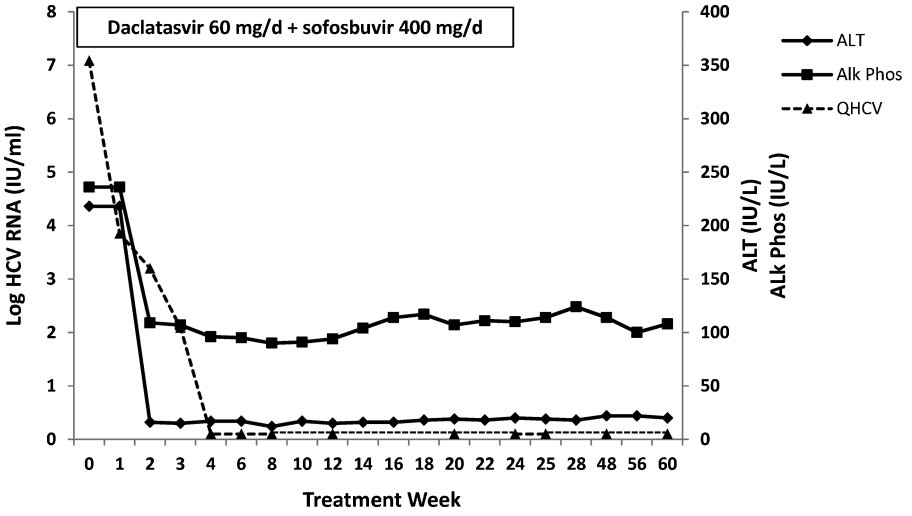

Figure 2: Serum HCV RNA was 12 000 000 IU/mL, ALT 218 IU/L and alkaline phosphatase 236 IU/L prior to treatment. Within 2 weeks of initiating combination therapy, the serum ALT and alkaline phosphatase levels had normalized while serum HCV RNA became undetectable (<43 IU/mL) at week 4 and remained so during and after treatment. The patient has remained HCV RNA negative with normal liver biochemistries at 9 months of follow-up consistent with a sustained virological response.

level of 4–6 ng/mL. During posttreatment follow-up, the patient did not receive any additional antiviral agent and was maintained on tacrolimus. He has remained well with undetectable HCV RNA at 9 months follow-up consistent with a sustained virological response. In addition, his serum AST 13 IU/mL, ALT 18 IU/mL, alk phos 101 IU/mL and total bilirubin of 0.5 mg/dL remain normal. Finally, his serum creatinine is stable at 1.3 mg/dL, hemoglobin 15.4 g/dL, WBC 7.8 $\times$ 10^3/mL and platelets of 195 $\times$ 10^3/mL.

Daclatasvir Pharmacokinetics

Daclatasvir pharmacokinetic parameters were stable throughout dosing (Table 1). The daclatasvir trough concentrations ranged from 57 to 196 ng/mL which is within the range of exposure where DCV has demonstrated antiviral activity when co-administered alone or in combination with peginterferon and ribavirin or other direct acting antiviral agents. The DCV T_{max} for this subject was 4 h with a C_{max} of 597 ng/mL which are also within the anticipated range of exposure.

Table 1: Daclatasvir Pharmacokinetic parameters during 24 weeks of treatment with sofosbuvir

Week 2 pK data (time in hours)	Daclatasvir concentration (ng/mL)
0	57
0.5 h	46
1.0 h	48
2.0 h	215
4.0 h	597
6.0 h	400
Study week	**AM trough concentration (ng/mL)**
6	76
8	91
12	149
24	196

Discussion

Severe recurrent HCV infection following LT is very challenging to manage. Interferon and ribavirin are difficult to initiate in many LT recipients due to their persistent pancytopenia from residual portal hypertension/splenomegaly and chronic renal insufficiency that precludes use of full dose peginterferon and ribavirin, respectively (4). In addition, as many as 30–70% of treated patients require a medication dose reduction and 20–40% terminate therapy prematurely due to intolerable side effects. Furthermore, there is increasing recognition of immune-mediated allograft dysfunction due to Interferon that may not only require early discontinuation of treatment but also lead to premature graft failure and/or even death (5,17). However, since recent studies have demonstrated that LT recipients who achieve SVR have a significantly improved survival compared to nonresponders and untreated patients, there is an urgent need to develop safer and more effective therapies for LT recipients (18,19).

The current study demonstrates that sofosbuvir in combination with daclatasvir is an efficacious and well-tolerated interferon free regimen in a LT recipient with severe recurrent cholestatic HCV infection. Ongoing studies have demonstrated that each of these agents when combined

with peginterferon and ribavirin markedly improves SVR rates compared to peginterferon and ribavirin alone in HCV genotype 1 patients (11,13). In addition, potent antiviral efficacy of sofosbuvir combined with DCV with or without ribavirin for 24 weeks has been demonstrated in HCV genotype 1 patients (20). In those phase 2 studies, 100% of patients became HCV RNA negative at week 4 and there was no evidence of resistance to either agent during or after therapy. Consistent with these findings, our interferon refractory patient who had a very high baseline HCV RNA level in the setting of tacrolimus and prednisone immunosuppression, achieved a serum HCV RNA below the lower limit of detection at week 4 and remained undetectable during and after treatment. These data demonstrate the potent antiviral activity of these individual agents when coadministered even to an immunosuppressed LT recipient. Furthermore, the excellent safety and side effect profile of this drug combination makes this interferon free regimen an attractive treatment option for other LT recipients with recurrent HCV infection. Daclatasvir in combination with peginterferon and ribavirin has generally been well-tolerated in over 1100 patients treated worldwide. No significant increase in the rate of laboratory or clinical adverse events was observed when compared to peginterferon and ribavirin alone and no unique daclatasvir related adverse events were identified (21). Sofosbuvir has also been generally well tolerated with use in over 500 patients worldwide and an adverse event profile similar to placebo when given with ribavirin alone or in combination with peginterferon and ribavirin (22). However, the dose and/or dosing interval of sofosbuvir may need to be altered in patients with severe renal impairment (i.e. GFR $<$ 30 mL/min) as renal elimination is the main route of clearance for sofosbuvir and its metabolites (23). In ongoing studies, some patients receiving sofosbuvir have reported headache and insomnia which are difficult to attribute to the sofosbuvir versus the peginterferon and ribavirin. Although preliminary data demonstrate that boceprevir or telaprevir with peginterferon and ribavirin can lead to rapid suppression of HCV replication in LT recipients with genotype 1, significant side-effects including transfusion dependent anemia, renal insufficiency, infection and even death have been reported highlighting the need for simpler and better tolerated therapies (8,9).

The lack of clinically apparent drug–drug interactions with the calcineurin inhibitors make both daclatasvir and sofosbuvir very attractive for future use in LT recipients with recurrent HCV infection (14,15). Of note, another LT recipient with severe recurrent HCV infection was recently treated with DCV, peginterferon and ribavirin for 24 weeks and did not experience any apparent drug–drug interactions with cyclosporine (24). In conclusion, the current study demonstrates the feasibility of combining potent oral antiviral agents with differing mechanisms of action in an LT recipient with severe recurrent cholestatic HCV infection. Although this African American patient had an unfavorable IL28-B genotype and a lack of response to prior peginterferon and ribavirin therapy, he rapidly responded to this interferon-free regimen. This case illustrates that the use of potent oral antiviral agents such as daclatasvir and sofosbuvir even early after transplantation offers great promise to the many HCV patients worldwide who are experiencing reduced quality of life and survival due to recurrent infection (2,18). Based upon this case, prospective studies involving these and other investigational oral antiviral agents in development should be undertaken for the growing number of LT recipients with clinically significant recurrent HCV infection. In addition, use of interferon-free regimens is of great interest in LT candidates with decompensated cirrhosis to reduce the frequency and severity of HCV recurrence but further studies in this difficult to treat population are needed.

Acknowledgments

This work was supported in part by the NIH through its support of the Michigan Institute for Clinical and Health Research (UL1RR024986). The authors would like to thank the study coordinators Kristin Chesney, Sonal Trivedi and Suzanne Welch as well as the research staff at MICHR for their assistance in the conduct of this study.

Disclosure

The authors of this manuscript have conflicts of interest to disclose as described by the *American Journal of Transplantation*. Eric Hughes, Marc Bifano and Dessie Dimitrova are employees of Bristol-Myers Squibb. William Symonds is an employee of Gilead Sciences and Robert Hindes is a former employee of Gilead Sciences. Dr. Fontana is conducting other research protocols with Gilead and Bristol-Meyers Squibb. Dr. Appelman has no potential conflicts.

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