JGH Journal of Gastroenterology and Hepatology

JGHF

doi:10.1111/jgh.12029

REVIEW

A pilot study of umbilical cord-derived mesenchymal stem cell transfusion in patients with primary biliary cirrhosis

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Key words

primary biliary cirrhosis, ursodeoxycholic acid, mesenchymal stem cell, immune regulation.

Abbreviations

ALB, albumin; ALP, alkaline phosphatase; AST, aspartate aminotransferase; CD, cluster of differentiation; CRE, creatinine; GGT, γ-glutamyltransferase; INR, international normalized ratio; MELD, model for end-stage liver disease; MRS, Mayo risk score; MSC, mesenchymal stem cells; PBC, primary biliary cirrhosis; PTA, prothrombin time activity; TBil, total bilirubin; UDCA, ursodeoxycholic acid; UC-MSC, umbilical cord-derived mesenchymal stem cell.

Accepted for publication 29 October 2012.

Abstract

Background and Aim: Ursodeoxycholic acid (UDCA) treatment is an effective medical therapy for patients with primary biliary cirrhosis (PBC); however, 40% of PBC patients show an incomplete response to the UDCA therapy. This study aimed to investigate the safety and efficacy of umbilical cord-derived mesenchymal stem cell (UC-MSC) transfusion in PBC patients with an incomplete response to UDCA.

Methods: We conducted a single-arm trial that included seven PBC patients with a suboptimal response to UDCA treatment. UC-MSCs were first cultured, and then 0.5×10^6 cells/kg body weights were infused through a peripheral vein. UC-MSCs were given three times at 4-week intervals, and patients were followed up for 48 weeks. Primary outcomes were to evaluate the safety and feasibility of UC-MSC treatment, and secondary outcomes were to evaluate liver functions and patient's quality of life.

Results: No obvious side-effects were found in the patients treated with UC-MSCs. Symptoms such as fatigue and pruritus were obviously alleviated in most patients after UC-MSC treatment. There was a significant decrease in serum alkaline phosphatase and γ-glutamyltransferase levels at the end of the follow-up period as compared with baseline. No significant changes were observed in serum alanine aminotransferase, aspartate aminotransferase, total bilirubin, albumin, prothrombin time activity, international normalized ratio, or immunoglobulin M levels. The Mayo risk score, a prognostic index, was also stable during the treatment and follow-up period.

Conclusions: UC-MSC transfusion is feasible and well tolerated in patients with PBC who respond only partially to UDCA treatment, thus representing a novel therapeutic approach for patients in this subgroup. A larger, randomized controlled cohort study is warranted to confirm the clinical efficacy of UC-MSC transfusion.

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Introduction

Primary biliary cirrhosis (PBC) is a progressive autoimmune liver disease that causes substantial loss of intrahepatic bile ducts, ultimately resulting in cholestasis, advanced fibrosis, cirrhosis, liver failure, and even hepatocellular carcinoma. Ursodeoxycholic acid (UDCA) is currently the only drug specifically approved for the treatment of PBC.[1] Patients who respond to UDCA treatment have a life expectancy comparable with the general population;[2] however, more than 40% of patients have an incomplete response to UDCA, resulting in progressive disease necessitating liver transplantation or death from liver-related causes.[3]

Currently, no efficient treatment is clinically available for this population of UDCA-resistant patients. Several clinical trials using different therapeutic agents to treat PBC patients with an incomplete response to UDCA, including silymarin,[4] oral

budesonide,[5] atorvastatin,[6] methotrexate[7] and rituximab,[8,9] have either failed to show significant clinical benefit or have some unacceptable safety profiles. Therefore, a new strategy to delay or prevent disease progression in PBC patients with an incomplete response to UDCA is urgently required.

Mesenchymal stem cells (MSCs) are multipotent non-hematopoietic progenitor cells capable of differentiating into multiple lineages.[10–13] MSCs have been used as a therapeutic strategy for tissue regeneration and repair, and their potential immunomodulatory capacity has also raised significant clinical interest.[14–17] Although these properties are not completely understood, emerging evidence from animal and human studies makes MSCs a promising therapeutic tool for autoimmune disease. The umbilical cord-derived MSC (UC-MSC) is of particular interest because of its relatively easy accessibility and abundant source,[18] making it a good substitute for MSC in future clinical studies.

Recently, transfusion of UC-MSCs has been reported to significantly improve symptoms in patients with severe autoimmune diseases, such as severe and refractory systemic lupus erythematosus,[19] therapy-resistant rheumatoid arthritis,[20] and immune thrombocytopenia patients,[21] with few adverse effects. Recently, our own research has indicated that UC-MSC therapy is well tolerated and has the potential to improve liver function, and reduce ascites and mortality in hepatitis B virus-associated patients with decompensated liver cirrhosis[22] and liver failure,[23] respectively. The goal of the present pilot study was to evaluate the safety and initial efficacy of UC-MSC transplantation in PBC patients with an incomplete response to UDCA therapy.

Materials and methods

Patients. Seven PBC patients with an incomplete response to UDCA were enrolled in the study between May 6, 2010 and March 5, 2011 in Research Center for Biological Therapy/Beijing 302 Hospital. These patients (ages between 33 and 58 years) were diagnosed with PBC based on the presence of an antimitochondrial antibody (AMA) titer $> 1:40$, and serum alkaline phosphatase (ALP) at least twice the upper limit of normal in the absence of biliary obstruction, which was in accordance with the American Association for the Study of Liver Diseases practice guidelines.[1] Additionally, enrolled patients did not have a normalization of their ALP after a minimum of six months of treatment with adequate doses of UDCA.[8,24,25] The exclusion criteria were as follows: pregnancy; coexisting liver disease (hepatitis A, hepatitis B, and hepatitis C, etc.); vital organ failure (cardiac, renal, or respiratory); the presence of any underlying neoplasm; the presence of hepatic, portal, or splenic vein thromboses on Doppler ultrasonography; treatment with immunosuppressive medication or any experimental drug within six months of enrollment; evidence of extrahepatic biliary disease; active substance abuse; lack of a supportive family; and if the patients were unwilling to sign the informed consent form.

This study was registered at ClinicalTrials.gov of the National Institutes of Health of the USA (registration number: NCT01662973) and was authorized by the General Logistic Ministry of Health, China. After the approval of the project by the Ethics Committee of Beijing 302 Hospital, all patients signed a written informed consent form in accordance with the Institutional Review Board guidelines for the protection of human subjects. Mayo risk score (MRS) for patient 2 suggested that he is the most optimal candidate for liver transplantation, but there is no matched liver donor for him, therefore he received UC-MSC treatment.

Preparation, identification, and transfusion of UC-MSC. UC-MSCs were prepared, identified, and transfused according to our recently published protocol.[22] In brief, the mesenchymal tissues from umbilical cord vessels were diced into cubes, washed, and finally seeded into a T75-cm^2 tissue culture flask. The fourth passages of UC-MSCs were used for clinical transfusion into patients. Before transfusion, UC-MSCs were subjected to quality control, including the detection of CD31, CD34, CD105, CD45, CD90, CD29, CD44, CD73, and human leukocyte antigens-D region (HLA-DR), ALP, and oil red O staining, as well as bacteriological testing. Cells were then suspended at a concentration of 0.5×10^6 cells/kg body weight in saline and were slowly infused intravenously. Each patient received a UC-MSC transfusion once every four weeks on three occasions and was then followed up for an additional 40 weeks (Fig. 1). During the treatment and follow-up period, patients were also simultaneously given traditional UDCA therapy.

Follow-up and outcome measures. PBC patients with an incomplete response to UDCA were treated with UC-MSCs transfusions in combination with standard UDCA therapy. The following tests were performed at week 0, 24, and 48 after the onset of UC-MSC treatment. At each visit, a general physical examination and laboratory studies were carried out, including: liver function tests for serum total bilirubin (TBil), albumin (ALB), ALP, aspartate aminotransferase (AST), alanine aminotransferase, γ-glutamyltransferase (GGT), immunoglobulin A (IgA), immunoglobulin G (IgG), immunoglobulin M (IgM), total cholesterol, and α-fetoprotein (AFP); renal function markers including urea, uric acid (UA), and creatinine (CRE); thrombin markers including prothrombin time activity (PTA), and international normalized ratio (INR); and routine blood tests including white blood cell count and hemoglobin and platelet counts. The volume of hypogastric ascites was determined by ultrasonography, and the MRS[26] and model for end-stage liver disease (MELD) score were used to evaluate prognosis. History taking and physical examinations were also performed at each clinical visit. The presence of fatigue, pruritus, fever, peripheral edema, rash, nausea, vomiting, and other complications were recorded in detail at each visit.

MRS is calculated using the following equitation: $= 0.871 \times \log e$ (bilirubin[mg/dL]) $- 2.53 \times \log e$ (ALB [g/dL]) $+ 0.039 \times$ age(years) $+ 2.38 \times \log e$(prothrombin time [s]) $+ 0.859$ edema (0 = no edema, no diuretic therapy; 0.5 = edema, no diuretic therapy or no edema, diuretic therapy; 1 = edema and diuretic therapy); MELD score is calculated using the following equation: $= 3.8 \times \log e$[bilirubin(mg/dL)] $+ 11.2 \times \log e$(INR) $+$ $9.6 \times \log e$[CRE(mg/dL)] $+ 6.4 \times$ (biliary or alcoholic = 0; others = 1).

Statistical analysis. Statistical analyses were performed using SPSS statistical software (version 13.0; SPSS Inc., Chicago,

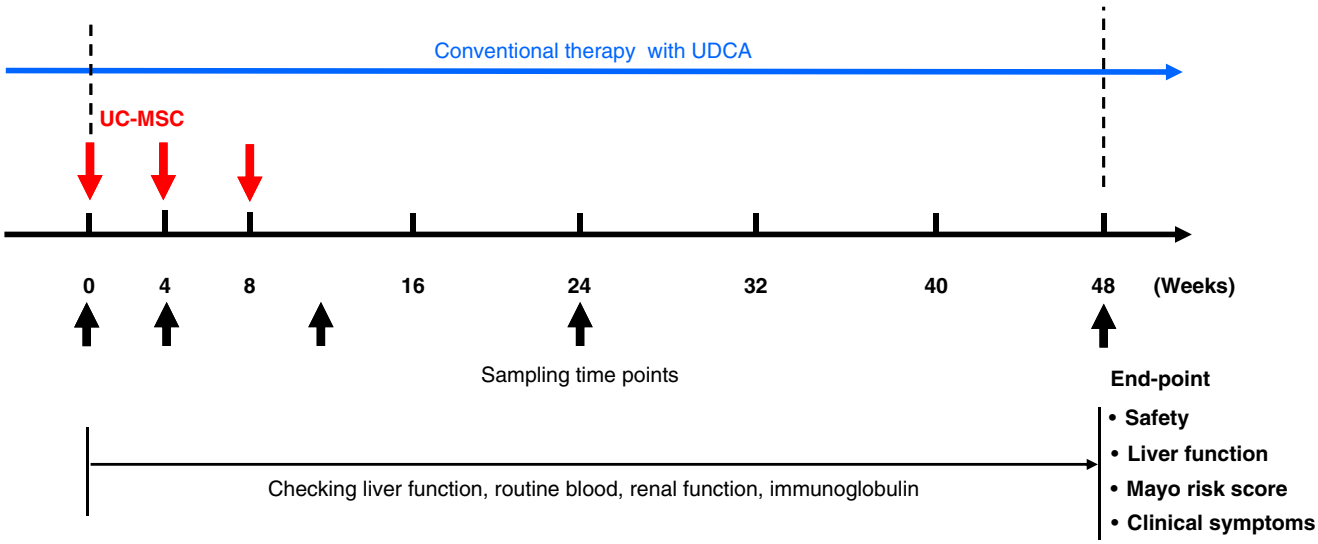

Figure 1 Protocol for umbilical cord-derived mesenchymal stem cell (UC-MSC) treatment in primary biliary cirrhosis (PBC) patients with an incomplete response to ursodeoxycholic acid (UDCA). UC-MSC transfusions were given to the patients three times at the baseline (0-week), 4-week, and 8-week time points. The clinical parameters were tested at the 0-, 24-, and 48-week time points during the follow-up period.

Table 1 Baseline characteristics of enrolled patients

Characteristic	Patient 1	Patient 2	Patient 3	Patient 4	Patient 5	Patient 6	Patient 7
Age (years)	58	46	49	54	33	46	57
Gender (M/F)	F	M	F	F	F	F	F
History PBC (years)	6	6	6	7	7	2	12
AMA	Positive	Positive	Positive	Positive	Positive	Positive	Positive
Ascites	None	None	Mild	Mild	Minimal	None	Minimal
Presence of fatigue	+	+	+	+	+	+	+
Presence of pruritus	+	–	+	–	+	+	+
UDCA treatment start date	December 2008	December 2006	April 2010	March 2010	June 2005	June 2010	December 2002

All patients have an incomplete response to ursodeoxycholic acid (UDCA).
AMA, antimitochondrial antibody; PBC, primary biliary cirrhosis.

IL). The two-tailed Wilcoxon matched pairs test was used to evaluate paired biochemical values. Values with $P < 0.05$ were considered statistically significant.

Results

Patient characteristics. The demographic and clinical characteristics of patients at the time of enrollment are summarized in Table 1; this table also indicates when patients begun UDCA therapy. These patients included one male and six females, ranging from 33 to 58 years of age. All of the patients had been on UDCA therapy for at least one year and had shown an incomplete response to this treatment. All of the patients had reported fatigue, and five of them had pruritus at the beginning of UC-MSC therapy. Patient medication was not changed during the study period. The timeline of UC-MSC treatment in this group of patients is shown in Figure 1; note that clinical parameters were tested at the 0-, 24-, and 48-week time points during the study period.

Safety of UC-MSC transfusion. Because safety is a major concern regarding UC-MSC therapy for PBC patients, we examined the short-term side-effects and long-term adverse events during the 48 weeks of follow-up. All the seven patients tolerated the UC-MSC treatment well; only one patient developed a self-limiting fever (body temperature: 37–38°C) within 5 h of UC-MSC transfusion, which recovered within 12 h without any additional treatments. No short-term clinical adverse effects, such as right upper quadrant pain, skin rash, infection, coma, or shock, were reported. There were no occurrences of long-term complications, such as hepatocellular carcinoma, upper gastrointestinal hemorrhage, hepatic encephalopathy, or primary peritonitis within one year of follow-up.

Additionally, in all the seven patients, no significant alterations were observed in renal function parameters such as urea, CRE, and UA levels during the one-year follow-up period. Routine blood tests (peripheral white blood counts, hemoglobin, platelet, etc.), serum electrolyte levels (serum Na, serum K and serum Cl, etc.), and serum AFP levels also remained stable (Table 2).

Table 2 Laboratory parameters of the enrolled patients at baseline and during follow-up check points (routine blood, renal function and serum immunoglobulin)

Parameter	Patient 1			Patient 2			Patient 3			Patient 4			Patient 5			Patient 6			Patient 7		
	0 W	24 W	48 W	0 W	24 W	48 W	0 W	24 W	48 W	0 W	24 W	48 W	0 W	24 W	48 W	0 W	24 W	48 W	0 W	24 W	48 W
WBC (10^9/L)	2.64	3.02	2.85	4.05	6.13	4.58	3.35	4.52	3.10	4.50	2.89	3.30	4.46	4.78	4.80	4.89	5.26	4.61	4.37	6.16	6.88
NE (10^9/L)	1.31	1.25	0.89	2.74	4.25	3.14	1.93	2.86	2.15	2.84	1.80	1.46	3.12	3.34	3.50	3.07	3.34	2.75	1.40	2.96	3.74
LY (10^9/L)	0.96	1.24	0.71	0.83	1.20	1.08	1.10	1.32	0.78	1.32	0.85	1.37	0.88	1.02	0.91	1.32	1.48	1.52	2.07	1.80	1.56
MO (10^9/L)	0.24	0.38	0.20	0.25	0.34	0.25	0.24	0.22	0.13	0.33	0.23	0.46	0.32	0.25	0.25	0.37	0.32	0.27	0.81	1.26	1.47
EO (10^9/L)	0.12	0.14	0.05	0.23	0.33	0.11	0.06	0.11	0.03	0.00	0.00	0.00	0.11	0.15	0.12	0.10	0.09	0.00	0.07	0.13	0.07
BA (10^9/L)	0.01	0.01	0.00	0.00	0.01	0.00	0.02	0.01	0.01	0.01	0.01	0.01	0.03	0.02	0.02	0.03	0.03	0.02	0.02	0.01	0.04
RBC (10^{12}/L)	3.46	3.76	3.09	2.56	3.39	3.34	2.77	3.22	2.83	3.81	3.59	3.13	4.14	4.46	3.88	2.80	3.28	3.31	2.94	2.65	2.26
HGB (g/L)	109	117	98	89	115	111	60	86	68	116	106	96	105	107	88	86	101	101	98	91	84
PLT (10^9/L)	67	76	79	47	53	55	61	61	44	67	54	69	105	119	119	271	197	118	101	71	90
GLU (mmol/L)	4.13	4.18	3.81	4.57	4.97	4.60	4.02	4.54	10.87	5.24	4.00	3.90	5.27	5.30	5.41	5.82	5.12	5.04	3.68	4.37	5.00
TC (mmol/L)	6.26	6.00	3.76	1.79	3.91	4.45	2.60	2.78	3.05	4.50	3.69	2.98	7.44	5.01	1.86	1.91	2.01	2.25	2.70	2.52	1.63
Urea (mmol/L)	4.10	4.00	3.40	3.50	4.80	3.80	4.30	4.80	5.60	3.90	2.90	4.40	3.20	3.20	2.20	3.20	3.50	2.90	3.40	2.90	3.20
CRE (μmol/L)	46	55	42	89	77	69	61	60	62	63	65	71	79	65	46	51	56	60	61	63	69
UA (μmol/L)	213	283	196	119	111	115	183	188	164	193	207	298	165	214	158	167	186	177	256	339	274
Na (mmol/L)	144	143	138	134	139	140	139	139	139	137	142	141	144	144	141	145	140	141	143	140	132
K (mmol/L)	3.60	2.70	2.90	3.80	3.70	3.80	3.40	3.50	2.74	4.10	3.50	3.30	2.20	2.90	3.60	3.30	3.10	3.50	3.10	3.50	3.60
Cl (mmol/L)	105.2	99.5	103.8	107.4	103.5	105.8	111.5	109.2	115.6	107.5	108.2	104.2	101.1	109.5	105.1	106.5	101.2	104.3	104.6	106.5	94.9
IgA (g/L)	3.65	4.04	3.84	4.02	3.84	3.24	3.94	4.04	4.30	1.60	1.94	1.70	2.12	1.86	2.00	3.62	4.04	3.52	6.47	4.61	6.92
IgG (g/L)	25.13	27.65	28.15	23.78	23.48	20.34	30.31	28.89	34.45	12.31	11.89	13.26	20.53	16.81	17.98	14.11	16.47	13.70	21.09	13.14	14.85
IgM (g/L)	1.80	1.91	1.64	2.23	2.68	2.74	1.79	1.86	1.55	1.52	1.57	1.65	8.77	7.00	8.94	3.22	5.73	6.56	1.83	1.22	1.52
AFP (ng/mL)	12	16	10	9.3	17.3	6	8.4	8.4	10	8.4	5	9	8	12.2	21	10.7	13.8	9.3	5	6.8	17

AFP, α-fetoprotein; BA, basophil; Cl, chlorine; CRE, creatinine; EO, eosinophil; GLU, glucose; HGB, hemoglobin; IgA, immunoglobulin; IgG, immunoglobulin G; IgM, immunoglobulin M; K, potassium; LY, lymphocytes; MO, monocyte; Na, sodium; NE, neutrophil; PLT, platelet; RBC, red blood cell; TC, total cholesterol; UA, uric acid; W, week(s); WBC, white blood cell.

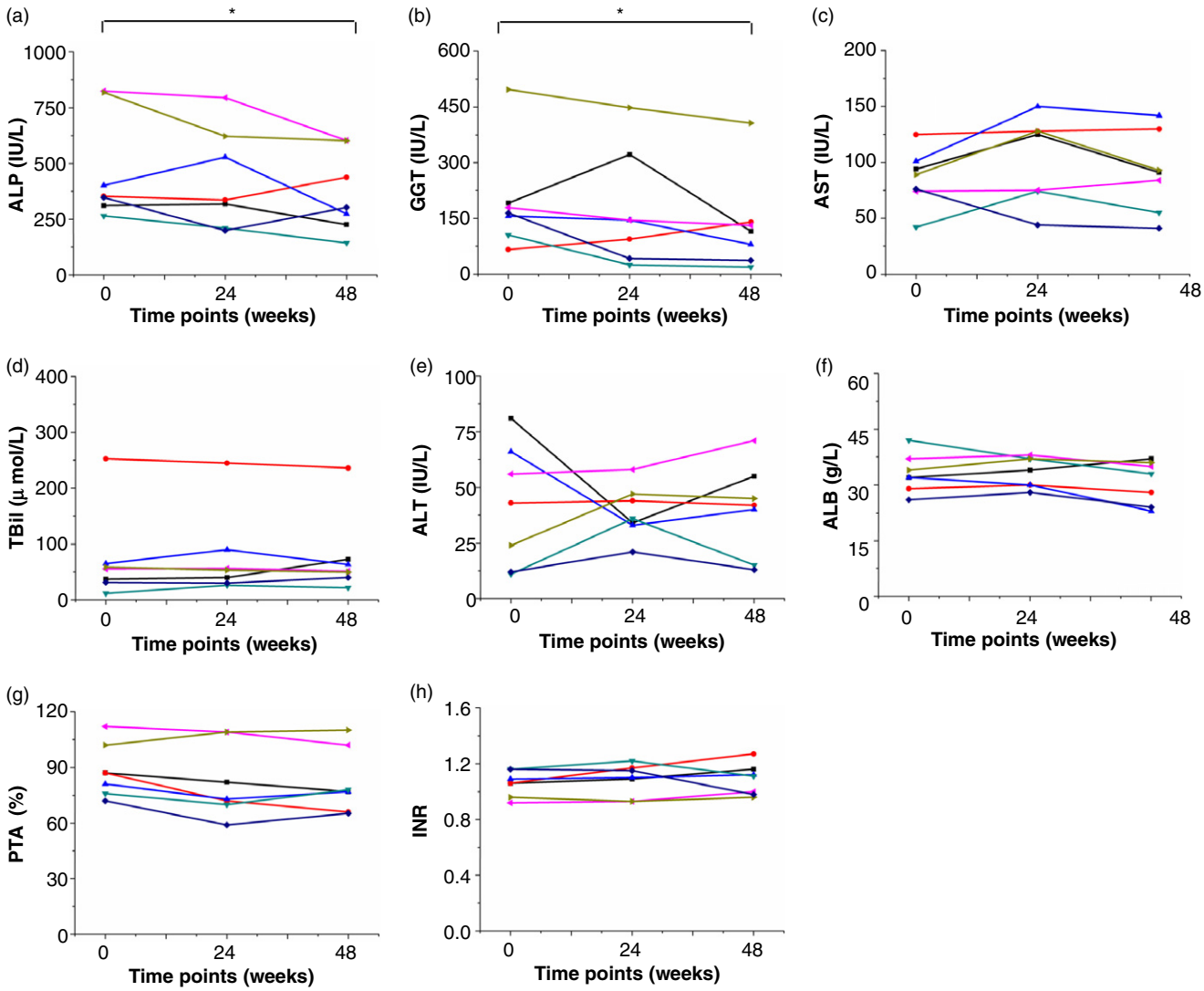

Figure 2 Umbilical cord-derived mesenchymal stem cell (UC-MSC) transfusions significantly decrease the serum alkaline phosphatase (ALP) and γ-glutamyltransferase (GGT) levels in primary biliary cirrhosis (PBC) patients with an incomplete response to ursodeoxycholic acid (UDCA). (a) Serum ALP, (b) GGT, (c) aspartate aminotransferase (AST), (d) total bilirubin (TBil), (e) alanine aminotransferase (ALT), (f) albumin (ALB), (g) prothrombin time activity (PTA), and (h) international normalized ratio (INR) were measured at 0 weeks (baseline), 24 weeks, and 48 weeks after the transfusion of UC-MSCs and continuing UDCA treatment. Each line represents the data from an individual patient. $*P < 0.05$ in two-tailed Wilcoxon matched pairs test. —, Pt 1; —, Pt 2; —, Pt 3; —, Pt 4; —, Pt 5; —, Pt 6; —, Pt 7.

Efficacy of UC-MSC transfusion in PBC patients: Liver function parameters. The combined treatment of UC-MSC and UDCA led to a significant decrease in serum ALP levels at the endpoint of the follow-up period (474.29 ± 223.26 *vs* 369.86 ± 168.35 IU/L, $P = 0.044$). Interestingly, GGT, another biochemical marker of cholestasis, also indicated a reduction from baseline (194 ± 140.65 IU/L) compared with the endpoint of the treatment (132.71 ± 129.4 IU/L, $P = 0.049$). However, no significant changes in serum ALP or GGT were observed at 24 weeks when compared with baseline data. In addition, no significant changes were noted in all other parameters including serum AST, TBil, ALB, PTA, INR, and IgM throughout the follow-up period (Fig. 2 and Table 2).

Efficacy of UC-MSC transfusion in PBC patients: MRS. The MRS was used to predict PBC patient survival.[27] In this study, there were increasing trends in the MRS for patients 1, 2, 4, and 7. This is of particular interest for patient 2, who displayed high levels of MRS but remained stable during the follow-up period. By contrast, no significant changes of MRS were found in patient 3, 5, and 6 during the follow-up period. Thus, when data from all the seven patients were analyzed, there was a statistically insignificant increase in MRS observed after treatment with UC-MSCs (3.47 ± 3.85) as compared with baseline data (2.98 ± 4.08; $P = 0.08$; Fig. 3a). In addition, no significant changes were found in MELD score throughout the whole process of this clinical trial (Fig. 3b).

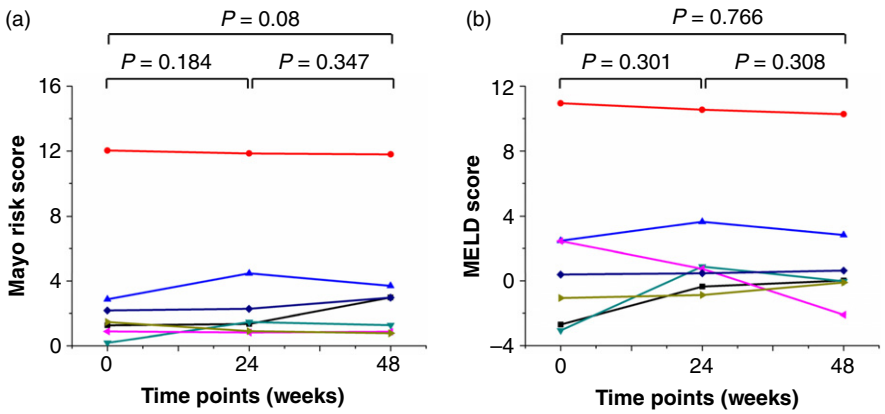

Figure 3 Mayo risk score (MRS) and model for end-stage liver disease (MELD) score are stable after the umbilical cord-derived mesenchymal stem cell (UC-MSC) transfusion. The (a) MRS and (b) MELD score were measured at 0 weeks (baseline), 24 weeks, and 48 weeks after the transfusion of UC-MSCs and continuing ursodeoxycholic acid (UDCA) treatment. Each line represents the data from an individual patient. —, Pt 1; —, Pt 2; —, Pt 3; —, Pt 4; —, Pt 5; —, Pt 6; —, Pt 7.

Efficacy of UC-MSC transfusion in PBC patients: Clinical symptoms. Fatigue and pruritus are common symptoms in patients with PBC. The symptom of fatigue is often described as perception of exhaustion resulting in a reduction of physical and mental capacity. We found that all the seven patients had fatigue to different extents before treatment, while at the end of the follow-up period, all the patients achieved subjective symptomatic alleviation of fatigue (Table 1). For patients 1, 3, 5, 6, and 7, their pruritus also underwent remission after 48 weeks of follow-up. In addition, we also found that the hypogastric ascites volumes of patients 3, 4, 5, and 7 were significantly decreased at week 24 and 48 since UC-MSC treatment (data not shown).

Discussion

PBC patients with an incomplete response to UDCA remain at increased risk for disease progression and represent a difficult-to-treat subpopulation. As such, a novel therapeutic regimen is urgently needed to treat these patients. UC-MSC transplantation, which has been shown to have a great impact on the symptoms of a variety of autoimmune diseases,[19,21] has been suggested by our group to be a potential new therapy to treat PBC patients with an incomplete response to UDCA therapy. In terms of its mechanism, UC-MSC treatment may lead to suppression of self-antigen-induced autoimmune conditions and facilitate repair of the injured bile duct caused by inflammation; however, the exact mechanism remains unknown and requires further study.

The present study indicated that UC-MSC transfusion through a peripheral vein is safe and feasible in PBC patients. No significant short-term side-effects or long-term complications were found during the study period. Similar to previous reports with regards to UC-MSC transfusion for other autoimmune diseases, UC-MSC treatment ameliorated some of the clinical symptoms in PBC patients and, therefore, may be clinically useful in the future. Importantly, this study suggests that UC-MSC treatment in PBC patients with an incomplete response to UDCA is clinically feasible and potentially efficacious. Serum ALP levels and MRS are two key parameters for the definition of response to treatment in patients with PBC; our data indicate that UC-MSC transfusion can significantly reduce the serum ALP levels and stabilize MRS during a 48-week follow-up period.

Our data also indicated that UC-MSC transfusion can improve the quality of life of patients with PBC. Fatigue and pruritus are the most common complaints from patients with PBC, which often affect the performance of daily activities and are significant contributors to impaired health-related life quality.[28,29] After UC-MSC treatment, all patients subjectively felt an improvement in their level of fatigue and pruritus at the endpoint of treatment. While this finding is promising, future studies should quantify fatigue and pruritus using more objective measures, such as the fatigue impact score[30] and the PBC-40 fatigue domain score[26] for fatigue, and the 5-D itch scale[31] and the visual analog scale[32] for pruritus.

Previous studies have shown that UC-MSC transfusion could reduce the titer of autoimmune antibodies in some types of autoimmune diseases. For example, Sun *et al.*[19] found that serum anti-double-stranded deoxyribonucleic acid antibodies became undetectable in some severe and refractory systemic lupus erythematosus patients after UC-MSC transfusion. Similarly, Ma *et al.*[21] demonstrated that UC-MSC treatment markedly suppressed IgG antiplatelet antibody secretion in immune thrombocytopenia patients. In our study, we assessed autoimmune parameters relevant to PBC, like AMA, IgA, IgM, and IgG; however, our results showed no obvious changes for all the detected parameters after treatment with UC-MSC and UDCA. Although we saw no change in serum AMA levels, it should be noted that there are some conflicting data on whether the serum level of AMA is correlated with the severity of PBC and if the AMA titer accurately predicts patient responses to treatment.[33,34] It is likely that modification of immune function following UC-MSC treatment will take a long time to show significant changes, as such, longer follow-up studies might be required to confirm the beneficial effect of UC-MSC treatment on these parameters.

Several limitations were present in this study. First, although some liver function and clinical symptoms were improved after UC-MSC transfusion, we cannot claim that such improvement is definitely related to UC-MSC transplantation in our small study

population of seven patients; rather, it may simply be related to the persistent UDCA treatment and/or natural course of PBC in these patients. The implementation of a larger randomized, controlled trial with a higher number of patients would clarify this issue. Second, we did not track the fate of UC-MSCs infused into our patients, which will be important to carry out in the future in order to understand the mechanism of UC-MSC function. Third, except for increasing bile flow and changing the hydrophobicity index of the bile acid pool,[35] UDCA could inhibit apoptosis, arrest cellular regeneration and block DNA repair in various cell types from hepatocytes to neurons.[36] UC-MSC transfusion in combination with UDCA will raise another concern whether UDCA will affect the function of UC-MSCs. Fourth, we did not document the histological alterations in the studied patients, which is the gold standard to evaluate treatment effects. Finally, there were only three time points for the follow-up study in this clinical trail, more detailed follow-up time points will be used in the future to provide an improved temporal resolution of changes in patient parameters during the follow-up period. Furthermore, the present study highlights several key issues that should be considered in future study designs, such as the minimum effective number of UC-MSCs to be administered, the optimal route of administration, and the optimal time for repeated therapy.

This study is the first to apply UC-MSC treatment in PBC patients. Our current findings demonstrate that UC-MSC transfusion via a peripheral vein is safe and yields promising results with regard to improved liver function and clinical symptoms in PBC patients with an incomplete response to UDCA treatment. Our results suggest that a large-scale, randomized, double-blinded, placebo-controlled clinical trial is warranted and should be conducted to confirm the use of UC-MSC treatment in this subgroup of PBC patients.

Acknowledgment

We greatly appreciate all the enrolled patients who participated in the clinical trial. This work was supported by grants from the Key Program of the National Ministry of Health and the PLA Grand Program on Clinical High and New Technology (Grant number: 200902002-2 and 2010gxjs098).

Conflict of interest

The authors have no conflicts of interest to declare.

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