Table 1. Baseline predictive factors before liver transplantation (pre-LT), at liver transplantation (at LT), and before interferon therapy (pre-IFN) associated with virorogical response (VR) and sustained VR (SVR): Univariate analysis.

		VR	non-VR	*P*	SVR	non-SVR	*P*
		n = 77	n = 31		n = 50	n = 67	
Age at LT (years)		55 (8–67)	56 (37–69)	0.462	54.5 (8–67)	56 (30–69)	0.212
Gender	Male	45 (74%)	16 (26%)	0.518	30 (46%)	35 (54%)	0.404
	Female	32 (68%)	15 (32%)		20 (38%)	32 (62%)	
HCC pre-LT	No	29 (71%)	12 (29%)	0.919	18 (43%)	24 (57%)	0.984
	Yes	48 (72%)	19 (28%)		32 (43%)	43 (57%)	
MELD pre-LT		15.5 (3–51)	15 (6–25)	0.403	16 (3–51)	15 (0–43)	0.616
Child-Pugh pre-LT	A/B	35 (74%)	12 (26%)	0.488	25 (49%)	26 (51%)	0.192
	C	41 (68%)	19 (32%)		24 (37%)	41 (63%)	
	unknown	1	0		1	0	
Serum HCV RNA pre-LT	<100 kIU/mL	16 (89%)	2 (11%)	0.063	11 (65%)	6 (35%)	0.028
	100 kIU/mL≤	52 (65%)	28 (35%)		31 (35%)	57 (65%)	
	unknown	9	1		8	4	
Serum HCV RNA pre-LT	<500 kIU/mL	50 (85%)	9 (15%)	<0.001	30 (55%)	25 (45%)	0.002
	500 kIU/mL≤	18 (46%)	21 (54%)		12 (24%)	38 (76%)	
	unknown	9	1		8	4	
Serum HCV RNA pre-LT	<1000 kIU/mL	56 (81%)	13 (19%)	<0.001	34 (49%)	36 (51%)	0.013
	1000 kIU/mL≤	12 (41%)	17 (59%)		8 (23%)	27 (77%)	
	unknown	9	1		8	4	
HCV genotype	Non-1	20 (100%)	0 (0%)	0.001	15 (79%)	4 (21%)	0.002
	1	57 (65%)	31 (35%)		35 (36%)	62 (64%)	
	unknown				0	1	
Donor age at LT (years)		42 (20–63)	38 (21–61)	0.504	43 (20–60)	38 (19–63)	0.748
Donor gender at LT	Male	41 (67%)	20 (33%)	0.287	27 (40%)	40 (60%)	0.538
	Female	36 (77%)	11 (23%)		23 (46%)	27 (54%)	
Sex mismatch	Match	28 (72%)	11 (28%)	0.932	18 (43%)	24 (57%)	0.984
	Mismatch	49 (71%)	20 (29%)		32 (43%)	43 (57%)	
ABO mismatch	Match	57 (66%)	29 (34%)	0.036	38 (40%)	56 (60%)	0.310
	Mismatch	20 (91%)	2 (9%)		12 (52%)	11 (48%)	
Relation of donor	Nonrelated	24 (73%)	9 (27%)	0.827	16 (44%)	20 (56%)	0.803
	Related	53 (71%)	22 (29%)		34 (42%)	47 (58%)	
Graft type	Left lobe	13 (81%)	3 (19%)	0.347	8 (62%)	5 (38%)	0.155
	Right lobe	64 (70%)	28 (30%)		42 (40%)	62 (60%)	
Splenectomy	No	38 (68%)	18 (32%)	0.413	25 (39%)	39 (61%)	0.378
	Yes	39 (75%)	13 (25%)		25 (47%)	28 (53%)	
Age pre-IFN (years)		57 (15–68)	57 (41–70)	0.494	56 (15–68)	57 (32–70)	0.200
Months from LT to therapy		9.2 (1.1–85.3)	8.9 (1.8–59.0)	0.846	9.0 (1.3–85.3)	9.0 (1.3–72.4)	0.879
Trough level for tacrolimus (ng/mL) pre-IFN		5.9 (2.0–10.9)	6.4 (3.3–10.6)	0.323	6.2 (2.2–9.5)	5.9 (2.0–12.7)	0.933
MMF pre-IFN	No	55 (71%)	23 (29%)	0.772	36 (43%)	48 (57%)	0.966
	Yes	22 (73%)	8 (27%)		14 (42%)	19 (58%)	
Prednisolone pre-IFN	No	64 (70%)	28 (30%)	0.347	41 (41%)	60 (59%)	0.245
	Yes	13 (81%)	3 (19%)		9 (56%)	7 (44%)	
Serum HCV RNA pre-IFN	<1000 kIU/mL	17 (89%)	2 (11%)	0.064	8 (38%)	13 (62%)	0.583
	1000 kIU/mL≤	58 (67%)	29 (33%)		42 (45%)	52 (55%)	
	unknown	2	0		0	2	
Serum HCV RNA pre-IFN	<5000 kIU/mL	52 (78%)	15 (22%)	0.020	36 (50%)	36 (50%)	0.030
	5000 kIU/mL≤	18 (55%)	15 (45%)		10 (28%)	26 (72%)	
	unknown	7	1		4	5	

Table 1. Cont.

		VR	non-VR	*P*	SVR	non-SVR	*P*
		n = 77	n = 31		n = 50	n = 67	
White cell count (102/mL)		51 (13–114)	49 (17–98)	0.135	49 (18–114)	48.5 (13–99)	0.049
Neutrophil count (102/mL)		26 (8–89)	22 (11–58)	0.127	26 (11–89)	23 (8–61)	0.044
Hemoglobin (g/dL)		12.0 (9.2–17.2)	12.0 (8.9–17.9)	0.638	12.0 (9.4–17.2)	11.8 (8.9–17.9)	0.157
Platelet count (104/mL)		21.7 (4.7–58.1)	15.1 (4.3–40.0)	0.153	20.3 (5.0–58.1)	15.8 (4.3–45.8)	0.165
AST (IU/L)		78 (19–352)	72 (25–464)	0.677	85 (21–352)	75 (24–547)	0.887
ALT (IU/L)		93 (18–395)	82 (21–392)	0.544	106 (22–395)	82 (18–597)	0.251
ALP (IU/L)		461 (199–1985)	433 (168–2977)	0.345	470 (204–1985)	470 (168–2977)	0.610
g-GTP (IU/L)		118.5 (15–1623)	114 (20–1827)	0.856	141 (15–1623)	115 (20–1827)	0.356
Bilirubin (mg/dL)		0.9 (0.3–11.0)	0.9 (0.3–10.4)	0.827	0.9 (0.4–11.0)	1.0 (0.3–13.7)	0.611
Activity grade pre-IFN	A1	54 (75%)	18 (25%)	0.448	35 (47%)	40 (53%)	0.517
	A2	22 (65%)	12 (35%)		14 (36%)	25 (64%)	
	A3	1 (50%)	1 (50%)		1 (33%)	2 (67%)	
Fibrosis stage pre-IFN	F0	9 (60%)	6 (40%)	0.446	6 (32%)	13 (68%)	0.530
	F1	54 (75%)	18 (25%)		34 (46%)	40 (54%)	
	F2/3	14 (67%)	7 (33%)		10 (42%)	14 (58%)	
Steatosis (5%<) pre-IFN	No	40 (69%)	18 (31%)	0.609	27 (42%)	38 (58%)	0.633
	Yes	36 (73%)	13 (27%)		23 (46%)	27 (54%)	
	unknown	1	0		0	2	
Cholestasis pre-IFN	No	58 (71%)	24 (29%)	0.903	38 (42%)	53 (58%)	0.577
	Yes	18 (72%)	7 (28%)		12 (48%)	13 (52%)	
	unknown	1	0		0	1	

NOTE. Qualitative variables are shown in number; and quantitative variables expressed as median (range). P-values are calculated by Wald test for logistic regression analysis.

LT, liver transplantation; HCC, hepatocellular carcinoma; MELD, model for end-stage liver disease; HCV, hepatitis C virus; MMF, mycophenolate mofetil; AST, aspartate aminotransferase; ALT, alanine aminotransferase; ALP, alkaline phosphatase; g-GTP, gamma-glutamyl transpeptidase.

doi:10.1371/journal.pone.0058380.t001

less than 500 kIU/mL, $P = 0.002$; and less than 1000 kIU/mL, $P = 0.013$), HCV genotype (non-1, $P = 0.002$), and low pretreatment serum HCV RNA levels (less than 5000 kIU/mL, $P = 0.030$). In addition, white cell count ($P = 0.049$) and neutrophil count ($P = 0.044$) before interferon therapy were significantly associated with SVR. Multivariate analysis showed that 2 variables were independently associated with SVR–a non-1 HCV genotype (OR: 0.182, 95% CI: 0.054–0.614, $P = 0.006$), and pretransplant serum HCV RNA levels lower than 500 kIU/mL (OR: 0.310, 95% CI: 0.130–0.742, $P = 0.009$) (Table 3). SVR rate among patients with a non-1 HCV genotype was 79% (15 of 19 patients) on average, 83% (10 of 12 patients) when pretransplant serum HCV-RNA level was less than 500 kIU/mL, and 50% (2 of 4 patients) when it was 500 kIU/mL or more. In patients with HCV genotype 1, SVR rate was 36% (35 of 97 patients) on average, 47% (20 of 43 patients) when pretransplant serum HCV-RNA level was less than 500 kIU/mL, and 22% (10 of 45 patients) when it was 500 kIU/mL or more.

Amino Acid Substitutions in Core Region of HCV

To determine the viral factors that predicted VR and SVR in patients infected with HCV genotype 1b, association of aa substitutions at aa 70 of arginine or glutamine/histidine and aa

Table 2. Predictive factors associated with virological response (VR): Multivariate analysis.

		Odds Ratio	95% confidence intervals	P-value
Serum HCV RNA pre-LT	<500 kIU/mL	1	-	-
	500 kIU/mL≤	0.178	0.054–0.535	0.001
HCV genotype	Non-1	1	-	-
	1	0.087	0.000–0.589	0.008
ABO mismatch	Match	1	-	-
	Mismatch	5.492	1.004-58.06	0.049

HCV, hepatitis C virus; LT, liver transplantation.

doi:10.1371/journal.pone.0058380.t002

Table 3. Predictive factors associated with sustained virological response (SVR): Multivariate analysis.

		Odds Ratio	95% confidence intervals	P-value
HCV genotype	Non-1	1	-	-
	1	0.182	0.054–0.614	0.006
Serum HCV RNA pre-LT	<500 kIU/mL	1	-	-
	500 kIU/mL≤	0.310	0.130–0.742	0.009

HCV, hepatitis C virus; LT, liver transplantation.
doi:10.1371/journal.pone.0058380.t003

91 of leucine or methionine with VR and SVR were analyzed in 40 patients, whose pre-treatment sera were stored (Table 4). As a result, substitutions of both aa 70 and aa 91 were not significantly associated with VR and SVR.

Predictors of Withdrawal from Therapy

Predictive factors for withdrawal from the treatment protocol were evaluated by comparing 26 patients who withdrew from the treatment protocol and the patients who completed the treatment including patients with SVR, patients who relapsed, and NR. None of the variables analyzed had a significant effect on withdrawal (Data not shown).

Discussion

In this study, we identified 2 independent predictors of SVR in patients with recurrent hepatitis C after LDLT by multivariate analysis: A non-1 HCV genotype and pretransplant serum HCV-RNA levels lower than 500 kIU/mL. The same factors were identified as predictors for VR, which purely indicates response to interferon therapy, by excluding the influences of the premature termination of the therapy and virological relapse after termination of the treatment. In addition, an ABO-incompatible LDLT was identified as an independent variable predicting VR.

In non-transplant settings, pretreatment predictors of response to interferon therapy have been analyzed in many studies, and the viral genotype and pretreatment viral load have been almost invariably shown to be 2 major predictors of SVR [41,42,43,44]. SVR rates were higher in patients infected with a non-1 HCV genotype and in those with a low pretreatment viral load. These 2 factors have been also identified in several reports [16,17,18,19] as factors predicting SVR in patients with recurrent hepatitis C after DDLT. In the present study, a non-1 HCV genotype was again identified as an independent predictive factor for both VR and SVR in patients with recurrent hepatitis C after LDLT by multivariate analysis. A pretreatment viral load <5000 kIU/mL was also a significant predictive factor by univariate analysis, but it was not an independently associated variable by multivariate analysis. On the other hand, pretransplant viral load was identified as an independent variable predictive of both VR and SVR by multivariate analysis.

While reports of factors that can control viral load exist, the mechanism by which serum HCV-RNA levels are regulated has not yet been completely clarified. A correlation between mutations in the ISDR sequence in the NS5A region of the HCV genome and serum HCV RNA levels has been reported. We did not analyze this viral factor in the current study; however, it is possible that the HCV genome sequence determines both pretransplant viremia and response to interferon therapy. The host polymorphism in IL28B, which was identified as a strong predictor of virological response to interferon therapy in patients with hepatitis C, was recently reported to be associated with baseline viral load [26,45]. The allele associated with a better treatment response is associated with a higher baseline viral load. This finding does not correspond with our results showing that a low HCV load predicts a better response to treatment. We speculate that the balance between host immunity and HCV replication regulates the serum HCV load, and that this balance also determines VR. As pretreatment viral load in post-transplant patients is influenced by immunosuppressive agents, the original host-virus balance

Table 4. Association of amino acid substitutions in the core region with virorogical response (VR) and sustained VR (SVR) in 40 patients infected with HCV genotype 1b: Univariate analysis.

		VR	non-VR	*P*	SVR	non-SVR	*P*
		n = 22	n = 13		n = 14	n = 24	
Core aa 70	Arg	9 (75%)	3 (25%)	0.289	7 (50%)	7 (50%)	0.204
	Gln/His	13 (57%)	10 (43%)		7 (29%)	17 (71%)	
Core aa 91	Leu	14 (64%)	8 (36%)	0.902	9 (38%)	15 (63%)	0.912
	Met	8 (62%)	5 (38%)		5 (36%)	9 (64%)	
Core aa 70 and 91	70 Arg and 91 Leu	6 (67%)	3 (33%)	0.784	5 (50%)	5 (50%)	0.320
	Others	16 (62%)	10 (38%)		9 (32%)	19 (68%)	
Core aa 70 and 91	70 Gln/His and 91 Met	5 (50%)	5 (50%)	0.324	3 (30%)	7 (70%)	0.603
	Others	17 (68%)	8 (32%)		11 (39%)	17 (61%)	

NOTE. Data are shown in number. P-values are calculated by Wald test for logistic regression analysis.
Arg, Arginine; Gln, glutamine; His, histidine; Leu, leucine; Met, methionine.
doi:10.1371/journal.pone.0058380.t004

would be reflected better by serum HCV levels before transplantation than by those after transplantation. It is unclear whether this result is specific to LDLT or holds true for both DDLT and LDLT. The significance of pretransplant viral load in DDLT as a predictor for virological response to post-transplant interferon therapy has not been analyzed in most previous studies [10]. Further analysis in patients who receive DDLT could help clarify the underlying mechanism.

Liver transplantation across the ABO blood-type barrier (ABO-incompatible) is generally contraindicated because of the possibility of graft loss caused by antibody-mediated rejection and is performed under exceptional circumstances as a rescue option in an emergent situation. However, ABO-incompatible LDLT has been performed in Japan to overcome organ shortage problems. Recently, rituximab prophylaxis and local infusion of prostaglandin E1 and steroids were established as therapeutic measures for recipients who underwent ABO-incompatible LDLT, and these treatments improved outcomes [46]. Interestingly, in this study, we found that an ABO-mismatched donor is associated with VR to interferon therapy. The reason for this interesting finding is unclear, but it is possible that either subclinical antibody-mediated rejection or drugs such as rituximab and prostaglandin E1 used in ABO-incompatible recipients may contribute to the higher VR to interferon therapy. There is hope that future studies to clarify the basic mechanism underlying this result will lead to a novel strategy to improve the efficacy of interferon therapy in patients with hepatitis C.

Amino acid substitutions of core region of HCV were not associated with treatment response in our analysis. We do not know the reason for the difference of impact of substitution of core aa 70 and aa 91 on virological response to interferon therapy from a previous report, in which SVR rate were significantly higher in transplant recipients with aa 70 of arginine and aa 91 of leucine of core region of HCV [33]. As sample size of both the previous study and our present study are small, and our present study did not assess the other HCV RNA mutations, including ISDR [32] and interferon/ribavirin resistance-determining region [47] in NS5A, and IL28B polymorphism in recipients and donors, further analysis should be required in larger cohorts.

Another aim of this study was to identify predictive variables for adverse events during interferon therapy, but none of the studied factors proved to be statistically significant predictors of withdrawal from the treatment protocol. As patients withdrew from the treatment for diverse reasons, it would be difficult to predict each adverse event before the initiation of interferon therapy. Therefore, careful follow-up during the treatment procedure is important for early detection of adverse events and to prevent progression to severe complications.

In this study, the final outcomes of the treatment including standard interferon plus ribavirin and peginterferon plus ribavirin were analyzed. Difference of the efficacy between standard interferon and peginterferon might affect the results of our present study. We predicted that patients who had virological response to standard interferon would also show the same response to peginterferon, because it is reported that the efficacy of peginterferon plus ribavirin is higher than that of standard interferon plus ribavirin [44,48]. Accordingly, the patients who achieved SVR by standard interferon were included in the present study. On the other hand, all nonresponders and all patients who relapsed by standard interferon plus ribavirin were retreated with peginterferon plus ribavirin, and we analyzed the final outcomes of the peginterferon plus ribavirin therapy. Therefore, we conclude that the difference of treatment regimen has little influence on our results.

In conclusion, SVR to antiviral therapy in patients with recurrent hepatitis C after LDLT is predictable before transplant by serum HCV-RNA level and HCV genotype. In addition, patients who undergo ABO-incompatible LDLT appear to have a better VR to interferon therapy after liver transplantation. Mechanisms underlying these interesting results are unknown at present, but these findings are likely to be useful for improved clinical assessment of patients with hepatitis C after liver transplantation, and could lead to development of new strategies for better outcomes in LDLT recipients with the HCV genotype 1 and/or a higher pretransplant viral load.

Author Contributions

Conceived and designed the experiments: YU HM. Performed the experiments: YU TK YO KO AY KH YF AMH HH HM. Analyzed the data: YU ST. Contributed reagents/materials/analysis tools: YU TK YO KO AY KH YF AMH HH HM. Wrote the paper: YU HM SU TC.

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Hepatology Research

Hepatology Research 2013; 43: 67–71 doi: 10.1111/j.1872-034X.2012.01020.x

Original Article

Efficacy and safety of prophylaxis with entecavir and hepatitis B immunoglobulin in preventing hepatitis B recurrence after living-donor liver transplantation

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Aim: Hepatitis B recurrence after liver transplantation can be reduced to less than 10% by combination therapy with lamivudine (LAM) and hepatitis B immunoglobulin (HBIG). The aim of this study was to evaluate the efficacy and safety of prophylaxis with entecavir (ETV), which has higher efficacy and lower resistance rates than LAM, combined with HBIG in preventing hepatitis B recurrence after living-donor liver transplantation (LDLT).

Methods: Twenty-six patients who received ETV plus HBIG (ETV group) after LDLT for hepatitis B virus (HBV)-related end-stage liver disease were analyzed by comparing with 63 control patients who had received LAM plus HBIG (LAM group).

Results: The survival rates of the patients treated with ETV plus HBIG was 73% after both 1 and 3 years, and there was no statistical difference between the patients in the ETV group and LAM group. No HBV recurrence was detected during the median follow-up period of 25.1 months in the ETV group, whereas the HBV recurrence rate was 4% at 3 years and 6% at 5 years in the LAM group. No patients had adverse effects related to ETV administration.

Conclusion: ETV combined with HBIG provides effective and safe prophylaxis in preventing hepatitis B recurrence after LDLT.

Key words: entecavir, hepatitis B, liver transplantation, living donor

INTRODUCTION

THE RECURRENCE OF hepatitis B virus (HBV) infection after liver transplantation for HBV-related diseases resulted in poor outcomes before the development of effective prophylaxis with lamivudine (LAM) and hepatitis B immunoglobulin (HBIG). Without the prophylaxis, the majority of patients developed recurrent infections due to HBV in the early phases after liver transplantation, and the recurrence resulted in rapidly progressive liver injury, early graft loss and reduced survival.[1–3] The development of prophylaxis dramatically reduced the post-transplant recurrence of hepatitis B and markedly improved prognosis. The most widely used prophylaxis so far has been a combination therapy of LAM and i.v. HBIG.

In the non-transplant setting, the long-term use of LAM resulted in high rates of emergence of resistance to the drug, with rates ranging 14–32% after 1 year and 60–70% after 5 years of treatment. In most cases, the resistance was the result of selection of LAM-resistant mutations in the YMDD motif of the DNA polymerase domain of HBV.[4] Moreover, the emergence of HBV strains with mutations that allow escape from hepatitis B surface antibody (anti-HBs) recognition has been reported in patients vaccinated for HBV,[5,6] in patients with chronic hepatitis B[7,8] and in liver transplant recipients after HBIG administration.[9–11] Therefore, the emergence of LAM resistance and HBIG resistance might

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Received 13 February 2012; revision 6 March 2012; accepted 28 March 2012.

increase the risk of recurrence during long-term administration of LAM and HBIG, although the rate of HBV recurrence in liver transplant recipients who received prophylaxis with LAM and HBIG for more than 10 years has not been reported to date. At present, several nucleoside analogs are available for the treatment of chronic hepatitis B[4]. Among them, there is entecavir (ETV), a carbocyclic analogue of 2′-deoxyguanosine, which has been shown to have higher efficacy than LAM in patients with chronic hepatitis B. In addition, ETV has a higher genetic barrier to resistance than LAM. The resistance to ETV requires at least three mutations including rtM204V/I, which causes LAM-resistance, rtL180M, and a mutation at one of the following codons: rtT184, rtS202 or rtM250.[4] Therefore, ETV is now used as a first-line therapy in the treatment of chronic hepatitis B worldwide. Data available in the published work suggest that, in transplant recipients, ETV plus HBIG represents a better prophylaxis protocol than LAM plus HBIG for long-term prevention of HBV recurrence after liver transplantation. However, the efficacy and safety of this treatment is largely unknown.

The aim of this study was to evaluate the efficacy and safety of prophylaxis with ETV and HBIG in preventing hepatitis B recurrence after living-donor liver transplantation (LDLT).

METHODS

Patients

WE RETROSPECTIVELY ANALYZED the medical records of 97 patients who underwent LDLT for HBV-related end-stage liver diseases from September 2002 to December 2010. Of these, eight patients were excluded from our study because they had breakthrough hepatitis due to HBV with LAM-resistant mutations and were prescribed LAM plus adefovir before liver transplantation. Accordingly, 89 patients were enrolled in this study.

Prophylaxis with ETV or LAM combined with HBIG

Lamivudine plus HBIG therapy was given to all recipients with HBV-related end-stage liver diseases from September 2002 to November 2006, as reported previously.[12] From December 2006, we changed the protocol for prophylaxis to ETV plus HBIG. ETV at a dose of 0.5 mg/day or LAM at a dose of 100 mg/day was given before transplantation, usually when the patient was referred to the hospital and scheduled for transplantation. Preoperative ETV or LAM prophylaxis was followed by combination with HBIG after transplantation. The first application of HBIG at a dose of 200 IU/kg body mass was administrated i.v. during the anhepatic phase of LDLT, and repeated every day for the first 5 days post-surgery. HBV serological markers were examined at weekly intervals for the first 2 months after the transplant, then at monthly intervals, and 1000 IU of HBIG was periodically administrated to maintain the serum anti-HBs titers at more than 500 IU/L during the first 6 months and 200 IU/L thereafter throughout the follow-up period.[12]

Immunosuppression

Tacrolimus and low-dose steroid therapy were administrated to induce immunosuppression in most patients.[13] Mycophenolate mofetil was administrated to patients who experienced refractory rejection or required reduction of tacrolimus dose due to adverse events. Patients who received ABO blood-type-incompatible transplants were treated with rituximab, plasma exchange, and hepatic artery or portal vein infusion with prostaglandin E1 and methylprednisolone.[14]

Diagnosis of HBV activation

Activation of HBV was diagnosed when hepatitis B surface antigens (HBsAg) and/or HBV DNA became positive in the serum of the patients. After LDLT, HBsAg, anti-HBs and serum HBV DNA were measured at least at 3 monthly intervals. Serological HBV markers, including HBsAg, anti-HBs, hepatitis B core antibody, hepatitis B e antigen (HBeAg) and antibodies to HBeAg (anti-HBe), were measured by chemiluminescent enzyme immunoassay (Fuji Rebio, Tokyo, Japan). Serum HBV DNA titer was analyzed using a commercial polymerase chain reaction (PCR) assay (Amplicor HBV Monitor; Roche, Branchburg, NJ, USA). LAM-resistant YMDD mutant virus was detected by the PCR enzyme-linked mini-sequence assay.[15]

Statistical analysis

Baseline characteristics are shown in Table 1. For continuous variables, medians and ranges are given, and the significance of the data was analyzed with the Wilcoxon rank sum test. For categorical variables, counts are given, and the data were analyzed with the χ^2-test. Survival rates and the rates of patients who showed HBV activation after LDLT were estimated using the Kaplan–Meier method and compared using log–rank tests. $P < 0.05$ was considered significant.

Table 1 Baseline characteristics of 90 patients

	Entecavir + HBIG ($n = 26$)	Lamivudine + HBIG ($n = 63$)	*P*-value
Age (years)	55 (33–68)	53 (26–64)	0.062†
Men/women	19/7	46/17	0.995‡
Primary disease			0.595‡
Acute liver failure	6 (23%)	9 (14%)	
Liver cirrhosis, HCC$^-$	6 (23%)	20 (32%)	
Liver cirrhosis, HCC$^+$	14 (54%)	34 (54%)	
HBV markers before LDLT			
HBsAg$^+$	24 (92%)	61 (97%)	0.350‡
HBeAg$^+$	6 (23%)	18 (29%)	0.595‡
HBV DNA before LDLT	<2.6 (<2.6–7.6<)	3.7 (<2.6–7.6<)	0.010†
<2.6 log IU/mL	14 (54%)	19 (30%)	0.024‡
Follow-up period (months)	25.1 (0.2–58.6)	70.6 (0.5–109.2)	<0.001†

Qualitative variables are shown in number; and quantitative variables expressed as median (range).
†Wilcoxon rank sum test.
‡χ^2-Test.
HBeAg, hepatitis B e antigen; HBIG, hepatitis B immunoglobulin; HBsAg, hepatitis B surface antigen; HBV, hepatitis B virus; HCC, hepatocellular carcinoma; LDLT, living-donor liver transplantation.

RESULTS

Patient characteristics

TWENTY-SIX PATIENTS who received ETV plus HBIG (ETV group) after LDLT for HBV-related end-stage liver disease were included in this study. Baseline characteristics of these patients are listed in Table 1 and compared with those of 63 control recipients who received LAM plus HBIG (LAM group) at our institute already present in our database. The two groups of patients did not differ significantly by age, sex, primary diseases or serological markers for HBV before LDLT. Serum HBV DNA levels before LDLT were significantly lower in the ETV group than in the LAM group. Fourteen of 26 patients (54%) showed less than 2.6 log IU/mL of serum HBV DNA in the ETV group. Median follow-up period was 25.1 months (range, 0.2–58.6) in the ETV group, whereas it was 70.6 months (range, 0.5–109.2) in the LAM group.

Efficacy and safety of prophylaxis with ETV plus HBIG

Survival rates of the patients treated with ETV plus HBIG estimated by Kaplan–Meier analysis was 73% at both 1 and 3 years (Fig. 1a). There was no difference between the ETV group and the LAM group, in which survival rates were 81% at 1 year, 78% at 3 years and 73% at

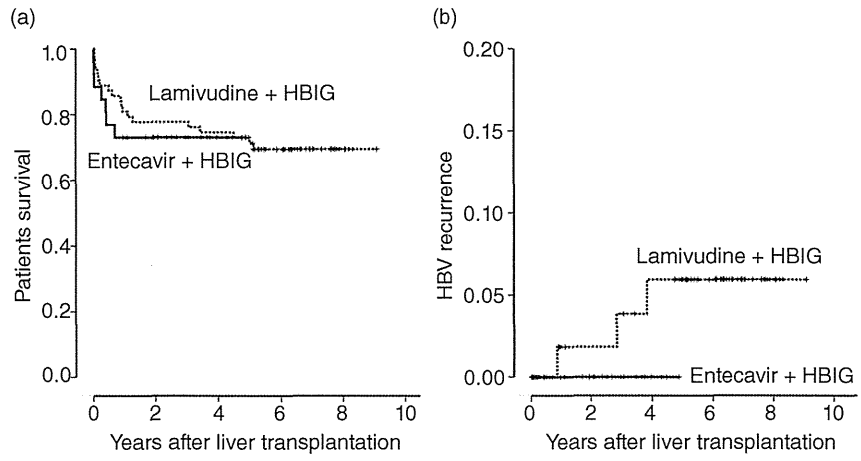

Figure 1 (a) Post-transplantation survival rates and (b) hepatitis B virus (HBV) recurrence after living-donor liver transplantation in HBV positive recipients who received entecavir and hepatitis B immunoglobulin (HBIG) (solid line), or lamivudine and HBIG (dotted line), estimated by Kaplan–Meier method.

5 years. Causes of death in patients in the ETV group were pneumonia ($n = 2$), sepsis ($n = 1$), pulmonary hemorrhage ($n = 1$), cerebral hemorrhage ($n = 1$), graft liver failure ($n = 1$) and multiple organ failure ($n = 1$), none of which were related to ETV. No HBV recurrence was detected in the median follow-up period of 25.1 months in the ETV group, whereas the HBV recurrence rate was 2% at 1 year, 4% at 3 years and 6% at 5 years in the LAM group (Fig. 1b). Three patients in the LAM group had HBV recurrence at 10, 34 and 46 months after LDLT. The emergence of HBV with LAM-resistant mutations in the YMDD motif was confirmed in two of the three patients. HBV mutations of another patient could not be determined because of the low level of serum HBV DNA. As the follow-up period of the ETV group was shorter than that of the LAM group and the HBV recurrence in the LAM group occurred in long-term follow-up after LDLT, the rate of HBV recurrence was not significantly different between the ETV and LAM groups. No patients had adverse events due to ETV administration.

DISCUSSION

IN THIS STUDY, we demonstrated that ETV combined with HBIG provides effective and safe prophylaxis in preventing hepatitis B recurrence after LDLT.

Two studies of patients receiving a combination of ETV and HBIG after liver transplantation have been previously reported.[16,17] One study demonstrated that 30 recipients who received ETV plus HBIG prophylaxis had no recurrence of HBV and no adverse effect relating to ETV.[17] The other study showed that no HBV recurrence was observed in two recipients with HBV-associated cirrhosis receiving ETV, tenofovir and HBIG.[16] Both studies showed the efficacy and safety of prophylaxis with ETV and HBIG in preventing short-term recurrence of HBV after liver transplantation. The current study confirmed their results for longer follow-up periods. Our results showed that prophylaxis with ETV and HBIG has similar efficacy and safety to that with LAM and HBIG, but did not show any further advantage of ETV compared to LAM treatment. Longer follow up might be needed to reveal the difference of HBV recurrence rate. One characteristic of our present report is that all patients in this study underwent LDLT. Our results suggest that prophylaxis with ETV and HBIG in patients after LDLT has similar efficacy and safety to patients after deceased-donor liver transplantation demonstrated in the previous reports.[16,17] More recently, efficacy of ETV monotherapy in preventing recurrence of HBV for liver transplant recipients with chronic hepatitis B was reported.[18] The study demonstrated that most patients showed disappearance of HBsAg and undetectable serum HBV DNA after liver transplantation without HBIG. Although long-term efficacy of ETV monotherapy needs be confirmed, both our data and previous reports suggest that ETV is an effective and safe antiviral agent in the post-transplant setting.

ACKNOWLEDGMENTS

THIS WORK WAS supported by Japan Society for the Promotion of Science (JSPS) Grants-in-Aid for Scientific Research (no. 21229009 and 23590972), Health and Labor Sciences Research Grants for Research on Intractable Diseases, and Research on Hepatitis from the Ministry of Health, Labor and Welfare, Japan, and a grant from Bristol-Myers-Squibb.

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