positive staining areas in 2 independent fields at 100× magnification using Lumina Vision 2.4 Bio-imaging software (Mitani Corporation, Tokyo, Japan). The following antibodies were used in this study: mouse anti-human AKR1B10 antibody (1:100 dilution; Ab 57547; Abcam, Cambridge, UK), anti-human HSP70 antibody (1:100 dilution; SC-24; Santa Cruz Biotechnology Inc., Santa Cruz, CA, USA), and anti-human GPC3 antibody (1:100 dilution; 1G12, Biomosaics, Burlington, VT, USA).

4.3. Statistical Analysis

All statistical analyses were performed using IBM SPSS 13.0 software (IBM SPSS, Chicago, IL, USA). Continuous variables were summarized as median (range), and Mann-Whitney *U*-tests or Kruskal-Wallis tests were used when appropriate. Univariate and multivariate regression analyses were used to examine the relationship of AKR1B10 expression in NT with demographic, histological, and biochemical variables. $p < 0.05$ was considered statistically significant.

5. Conclusions

We found that AKR1B10 expression is upregulated in chronic hepatitis or cirrhosis, preneoplastic conditions that predispose to HCC, in association with hepatic steatosis. Our findings could provide insight into the molecular mechanism of the very early stages of human hepatocarcinogenesis and a novel therapeutic target for the prevention of HCC.

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Conflicts of Interest

The authors declare no conflict of interest.

References

1. EI-Serag, H.B.; Rudolph, K.L. Hepatocellular carcinoma: Epidemiology and molecular carcinogenesis. *Gastroenterology* **2007**, *132*, 2557–2576.
2. Nakashima, O.; Sugihara, S.; Kage, M.; Kojiro, M. Pathomorphologic characteristics of small hepatocellular carcinoma: A special reference to small hepatocellular carcinoma with indistinct margins. *Hepatology* **1995**, *22*, 101–105.
3. Chuma, M.; Sakamoto, M.; Yamazaki, K.; Ohta, T.; Ohki, M.; Asaka, M.; Hirohashi, S. Expression profiling in multistage hepatocarcinogenesis: Identification of HSP70 as a molecular marker of early hepatocelllar carcinoma. *Hepatology* **2003**, *37*, 198–207.
4. Capurro, M.; Wanless, I.R.; Sherman, M.; Deboer, G.; Shi, W.; Miyoshi, E.; Films, J. Glypican-3: A novel serum and histochemical marker for hepatocellular carcinoma. *Gastroenterology* **2003**, *125*, 89–97.

5. Kondo, Y.; Kanai, Y.; Sakamoto, M.; Genda, T.; Mizokami, M.; Ueda, R.; Hirohashi, S. β-Catenin accumulation and mutation of exon 3 of the β-catenin gene in hepatocelllar carcinoma. *Jpn. J. Cancer Res.* **1999**, *90*, 1301–1309.
6. Oda, T.; Tsuda, H.; Scarpa, A.; Sakamoto, M.; Hirohashi, S. p53 gene mutation spectrum in hepatocellular carcinoma. *Cancer Res.* **1992**, *52*, 6358–6364.
7. Jin, Y.; Penning, T.M. Aldo-keto reductases and bioactivation/detoxication. *Annu. Rev. Pharmacol. Toxicol.* **2007**, *47*, 263–292.
8. Barski, O.A.; Tipparaju, S.M.; Bhatnagar, A. The aldo-keto reductase superfamily and its role in drug metabolism and detoxification. *Drug Metab. Rev.* **2008**, *40*, 553–624.
9. Scuric, Z.; Stain, S.C.; Anderson, W.F.; Hwang, J.J. New member of aldose reductase family proteins overexpressed in human hepatocellular carcinoma. *Hepatology* **1998**, *27*, 943–950.
10. Cao, D.; Fan, S.T.; Chung, S.S.M. Identification and characterization of a novel human aldose reductase-like gene. *J. Biol. Chem.* **1998**, *273*, 11429–11435.
11. Schmitz, K.J.; Sotiropoulos, G.C.; Baba, H.A.; Schmid, K.W.; Müller, D.; Paul, A.; Auer, T.; Gamerith, G.; Loeffler-Ragg, J. AKR1B10 expression is associated with less aggressive hepatocellular carcinoma: A clinicopathological study of 168 cases. *Liver Int.* **2011**, *31*, 810–816.
12. Teramoto, R.; Minagawa, H.; Honda, M.; Miyazaki, K.; Tabuse, Y.; Kamijo, K.; Ueda, T.; Kaneko, S. Protein expression profile characteristics to hepatocellular carcinoma revealed by 2D-DIGE with supervised learning. *Biochim. Biophys. Acta* **2008**, *1784*, 764–772.
13. Sato, S.; Genda, T.; Hirano, K.; Tsuzura, H.; Narita, Y.; Kanemitsu, Y.; Kikuchi, T.; Iijima, K.; Wada, R.; Ichida, T. Up-regulated aldo-keto reductase family 1 member B10 in chronic hepatitis C: Association with serum alpha-fetoprotein and hepatocellular carcinoma. *Liver Int.* **2012**, *32*, 1382–1390.
14. Sakamoto, M.; Effendi, K.; Masugi, Y. Molecular diagnosis of multistage hepatocarcinogenesis. *Jpn. J. Clin. Oncol.* **2010**, *40*, 891–896.
15. Fukumoto, S.; Yamauchi, N.; Moriguchi, H.; Hippo, Y.; Watanabe, A.; Shibara, J.; Taniguchi, H.; Ishikawa, S.; Ito, H.; Yamamoto, S.; *et al.* Overexpression of the aldo-keto reductase family protein AKR1B10 is highly correlated with smokers' non-small cell lung carcinomas. *Clin. Cancer Res.* **2005**, *11*, 1776–1785.
16. Li, C.-P.; Goto, A.; Watanabe, A.; Murata, K.; Ota, S.; Niki, T.; Aburatani, H.; Fukayama, M. AKR1B10 in usual interstitial pneumonia: expression in squamous metaplasia in association with smoking and lung cancer. *Pathol. Res. Pract.* **2008**, *204*, 295–304.
17. Wang, R.; Wang, G.; Richard, M.J.; Ferris, B.; Strulovici-Barel, Y.; Salit, J.; Hackett, N.R.; Gudas, L.J.; Crystal, R.G. Smoking-induced upregulation of AKR1B10 expression in the airway epithelium of healthy individuals. *Chest* **2010**, *138*, 1402–1410.
18. Trichopoulos, D.; Bamia, C.; Lagiou, P.; Fedirko, V.; Trepo, E.; Jenab, M.; Pischon, T.; Nöthlings, U.; Overevd, K.; Tjønneland, A.; *et al.* Hepatocellular carcinoma risk factors and disease burden in a European cohort: A nested case-control study. *J. Natl. Cancer Inst.* **2011**, *103*, 1686–1695.
19. Koh, W.-P.; Robien, K.; Wang, R.; Govindarajan, S.; Yuan, J.-M.; Yu, M.C. Smoking as an independent risk factor for hepatocellular carcinoma: The Singapore Chinese health study. *Br. J. Cancer* **2011**, *25*, 1430–1435.

20. Zeindl-Eberhart, E.; Haraida, S.; Liebmann, S.; Jungblut, P.R.; Lamer, S.; Mayer, D.; Jäger, G.; Chung, S.; Rabes, H.M. Detection and identification of tumor-associated protein variants in human hepatocellular carcinomas. *Hepatology* **2004**, *39*, 540–549.
21. Heringlake, S.; Hofdmann, M.; Fiebeler, A.; Manns, M.P.; Schmiegel, W.; Tannapfel, A. Identification and expression analysis of the aldo-ketoreductase1-B10 gene in primary malignant liver tumors. *J. Hepatol.* **2010**, *52*, 220–227.
22. Satow, R.; Shitashige, M.; Kanai, Y.; Takeshita, F.; Ojima, H.; Jigami, T.; Honda, K.; Kosuge, T.; Ochiya, T.; Hirohashi, S.; *et al.* Combined functional genome survey of therapeutic targets for hepatocellular carcinoma. *Clin. Cancer Res.* **2010**, *16*, 2518–2528.
23. Crosas, B.; Hyndman, D.J.; Gallego, O.; Martras, S.; Parés, X.; Flynn, T.G.; Farrés, J. Human aldose reductase and human intestine aldose reductase are efficient retinal reductases: consequences for retinoid metabolism. *Biochem. J.* **2003**, *373*, 973–979.
24. Gallego, O.; Ruiz, F.X.; Ardèvol, A.; Dominguez, M.; Alvarez, R.; de Lera, A.R.; Rovira, C.; Farrés, J.; Fita, I.; Parés, X.; Structural basis for the high all-trans-retinaldehyde reductase activity of the tumor marker AKR1B10. *Proc. Natl. Acad. Sci. USA* **2007**, *104*, 20764–20769.
25. Penning, T.M. AKR1B10: A new diagnostic marker of non-small cell lung carcinoma in smokers. *Clin. Cancer Res.* **2005**, *11*, 1687–1690.
26. Ruiz, F.X.; Porté, S.; Parés, X.; Farrés, J. Biological role of aldo-keto reductases in retinoic acid biosynthesis and signaling. *Front. Pharmacol.* **2012**, *3*, 58:1–58:42.
27. Wolbach, S.B.; Howe, P.R. Tissue changes following deprivation of fat-soluble A vitamin. *J. Exp. Med.* **1925**, *42*, 753–778.
28. Lancilloti, F.; Darwiche, N.; Celli, G.; de Luca, L.M. Retinoid status and the control of keratin expression and adhesion during the histogenesis of squamous metaplasia of tracheal epithelium. *Cancer Res.* **1992**, *52*, 6144–6152.
29. Darwiche, N.; Celli, G.; Sly, L.; Lancilloti, F.; de Luca, L.M. Retinoid status controls the appearance of reserve cells and keratin expression in mouse cervical epithelium. *Cancer Res.* **1993**, *53*, 2287–2299.
30. Schroder, E.W.; Rapaport, E.; Kabcenell, A.K.; Black, P.H. Growth inhibitory and stimulatory effects of retinoic acid on murine 3T3 cells. *Proc. Natl. Acad. Sci. USA* **1982**, *79*, 1549–1552.
31. Takatsuka, J.; Takahashi, N.; de Luca, L.M. Retinoic acid metabolism and inhibition of cell proliferation: An unexpected liaison. *Cancer Res.* **1996**, *56*, 675–678.
32. Tannenbaum, T.; Lowry, D.; Darwiche, N.; Morgan, D.L.; Gartsbein, M.; Hansen, L.; de Luca, L.M.; Hennings, H.; Yuspa, S.H. Topical retinoic acid reduces skin papilloma formation but resistant papillomas are at high risk for malignant conversion. *Cancer Res.* **1998**, *58*, 1435–1443.
33. Yan, R.; Zu, X.; Ma, J.; Liu, Z.; Adeyanju, M.; Cao, D. *Aldo-keto reductase family 1B10* gene silencing results in growth inhibition of colorectal cancer cells: Implication for cancer intervention. *Int. J. Cancer* **2007**, *121*, 2301–2306.
34. Muto, Y.; Moriwaki, H.; Saito, A. Prevention of second primary tumors by an acyclic retinoid, polyprenoic acid, in patients with hepatocellular carcinoma. *N. Engl. J. Med.* **1999**, *340*, 1046–1047.

35. Breton, J.; Gage, M.C.; Hay, A.W.; Keen, J.N.; Wild, C.P.; Donnellan, C.; Findlay, J.B.; Hardie, L.J. Proteomic screening of a cell line model of esophageal carcinogenesis identifies cathepsin D and aldo-keto reductase 1C2 and 1B10 dysregulation in Barrett's esophagus and esophageal adenocarcinoma. *J. Proteome Res.* **2008**, *7*, 1953–1962.
36. Pekow, J.R.; Bhan, A.K.; Zheng, H.; Chung, R.T. Hepatic steatosis is associated with increased frequency of hepatocellular carcinoma in patients with hepatitis C-related cirrhosis. *Cancer* **2007**, *109*, 2490–2496.
37. Hassan, M.M.; Hwang, L.Y.; Hatten, C.J.; Swaim, M.; Li, D.; Abbruzzese, J.L.; Beasley, P.; Patt, Y.Z.; Risk factors for hepatocellular carcinoma: Synergism of alcohol with viral hepatitis and diabetes mellitus. *Hepatology* **2002**, *36*, 1206–1213.
38. Bugianesi, E.; Leone, N.; Vanni, E.; Marchesini, G.; Brunello, F.; Carucci, P.; Musso, A.; de Paolis, P.; Capussotti, L.; Salizzoni, M.; *et al.* Expanding the natural history of nonalcoholic steatohepatitis: From cryptogenic cirrhosis to hepatocelllar carcinoma. *Gastroenterology* **2002**, *123*, 134–140.
39. Starmann, J.; Falth, M.; Spindelbock, W.; Lanz, K.-L.; Lackner, C.; Zatloukal, K.; Michael, T.; Sültmann, H. Gene expression profiling unravels cancer-related hepatic molecular signatures in steatohepatitis but not in steatosis. *PLoS One* **2012**, *7*, e46584.
40. Shaw, N.; Yang, B.; Millward, A.; Demaine, A. AKR1B10 is induced by hyperglycaemia and lipopolysaccharide in patients with diabetic nephropathy. *Cell Stress Chaperones* **2014**, *19*, 281–287.
41. Hirohashi, S.; Ishak, K.; Kojiro, M.; Wanless, I.; Theise, N.; Tsukuma, H.; Blum, H.; Deugnier, Y.; Puig, P.; Fischer, H.; *et al.* Tumors of the liver and intrahepatic bile ducts. In *Pathology and Genetics of Tumors of the Digestive System*; Hamilton, S.R., Aaltonen, L.A., Eds.; IARC Press: Lyon, France, 2000; pp. 157–202.
42. Desmet, V.J.; Gerber, M.; Hoofnagle, J.H.; Manns, M.; Scheuer, P.J. Classification of chronic hepatitis: Diagnosis, grading and staging. *Hepatology* **1994**, *19*, 1513–1520.
43. Tsuzura, H.; Genda, T.; Sato, S.; Hirano, K.; Kanemitsu, Y.; Narita, Y.; Kikuchi, T.; Iijima, K.; Wada, R.; Ichida, T. Association of visceral obesity with high viral load and histological findings in elderly patients with genotype 1 chronic hepatitis C. *Intern. Med.* **2013**, *52*, 1665–1673.

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REVIEW

Living-donor *vs* deceased-donor liver transplantation for patients with hepatocellular carcinoma

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Abstract

With the increasing prevalence of living-donor liver transplantation (LDLT) for patients with hepatocellular carcinoma (HCC), some authors have reported a potential increase in the HCC recurrence rates among LDLT recipients compared to deceased-donor liver transplantation (DDLT) recipients. The aim of this review is to encompass current opinions and clinical reports regarding differences in the outcome, especially the recurrence of HCC, between LDLT and DDLT. While some studies report impaired recurrence - free survival and increased recurrence rates among LDLT recipients, others, including large database studies, report comparable recurrence - free survival and recurrence rates between LDLT and DDLT. Studies supporting the increased recurrence in LDLT have linked graft regeneration to tumor progression, but we found no association between graft regeneration/initial graft volume and tumor recurrence among our 125 consecutive LDLTs for HCC cases. In the absence of a prospective study regarding the use of LDLT *vs* DDLT for HCC patients, there is no evidence to support the higher HCC recurrence after LDLT than DDLT, and LDLT remains a reasonable treatment option for HCC patients with cirrhosis.

Key words: Deceased donor liver transplantation; Hepatocellular carcinoma; Living donors; Living-donor liver transplantation; Recurrence

Core tip: The current opinions and clinical reports regarding differences in the recurrence of hepatocellular carcinoma (HCC) between living donor liver transplantation (LDLT) and deceased donor liver transplantation (DDLT) were reviewed. In the absence of a prospective study regarding the use of LDLT *vs* DDLT for HCC patients, only with some retrospective studies with conflicting results, there is no evidence to support the higher HCC recurrence after LDLT than DDLT, and LDLT remains a reasonable treatment option for HCC patients with cirrhosis.

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INTRODUCTION

Hepatocellular carcinoma (HCC) is the 7th most common cancer overall and the 3rd most common cause of cancer-related death worldwide[1,2]. Since the landmark report of the Milan criteria by Mazzaferro *et al*[3], which demonstrated comparable outcomes of patients with HCC having a single tumor smaller than 5 cm in diameter or up to 3 tumors smaller than 3 cm in diameter with no vascular invasion or extra-hepatic disease determined by preoperative imaging studies, deceased - donor liver transplantation (DDLT) has become an established treatment for cirrhotic patients with HCC[4,5]. Similarly, in Asian countries where living-donor liver transplantation (LDLT) comprises the majority of liver transplantation procedures, LDLT has become an established treatment

Table 1 Studies comparing living - donor liver transplantation and deceased - donor liver transplantation for hepatocellular carcinoma

Ref.	Country	Year	Study period	Type of LT	Case number	Recurrence - free survival			*P*	% Recurrence rate	*P*	Criteria used	% Outside Milan	Difference in tumor characteristics	Median follow-up period (mo)
						1-yr	3-yr	5-yr							
Impaired results in LDLT															
Park *et al*[10]	South Korea	2014	1999-2010	LDLT	166	89		81	0.045	19	0.045	UCSF	NA	none	35
				DDLT	50	96		94		6					
Vakili *et al*[13]	United States	2009	1999-2007	LDLT	28					29	< 0.05	UNOS	25	none	41
				DDLT	65					12					
Kulik *et al*[12]	United States	2012	1998-2010	LDLT	100	80	66	56	0.05	38	0.0004	UNOS	59	More aggressive in LDLT	60
	Multi-center			DDLT	97	90	81	73		11			30		
Lo *et al*[14]	Hong Kong	2007	1995-2004	LDLT	43	93	71	71	0.029	29	0.029	UCSF	26	More aggressive in LDLT	33
				DDLT	17	100	100	100		0			29		
Comparable results															
Sandhu *et al*[15]	Canada	2013	1996-2009	LDLT	58	88	75	70	NS	17	NS	Toronto criteria	28	none	38
				DDLT	287	86	75	70		15			32		31
Bhangui *et al*[16]	France	2011	2000-2009	LDLT	36	100	89	88	NS	13	NS	UCSF	27	none	58
				DDLT	120	93	89	86		13			21		50
Li *et al*[36]	China	2010	2005-2009	LDLT	38	71	42		NS	50	NS	UCSF	79	none	25
				DDLT	101	76	41			55			68		
Di Sandro *et al*[35]	Italy	2009	2000-2007	LDLT	25		96	96	NS	4	NS	Milan	20	none	NA
				DDLT	154		91	89		11			31		
Sotiropoulos *et al*[20]	Germany	2007	1998-2006	LDLT	45	88	75		NS	12	NS	UCSF	44	none	NA
				DDLT	55		81			14					
Hwang *et al*[8]	South Korea	2005	1992-2002	LDLT	237	83	80		NS	18	NS		27	none	26
	Multi-center			DDLT	75	88	82			16			29		45
Gondolesi *et al*[17]	United States	2004	1988-2002	LDLT	36	82	74		NS	19	NS	UNOS	53	none	15
				DDLT	165	90	83			19					

DDLT: Deceased - donor liver transplantation; HCC: Hepatocellular carcinoma; LDLT: Living - donor liver transplantation; LT: Liver transplantation; UCSF: University of California, San Francisco; UNOS: United Network for Organ Sharing; NA: Not applicable; NS: Not significant.

for HCC patients with end-stage liver disease[6,7]. LDLT is now considered a promising treatment for HCC patients in Western countries, not only to compensate for the shortage of donor organs but also to reduce the dropout rate on the waiting list[8].

With the accumulation of LDLTs for HCC patients, the impact of LDLT on recipient outcome compared with DDLT, especially the recurrence of HCC after liver transplantation, has become an important topic of debate[9]. The aim of this review was to encompass the current opinions and clinical reports regarding the differences in outcome, especially the recurrence of HCC, between LDLT and whole liver DDLT.

STUDIES COMPARING LDLT AND DDLT FOR HCC PATIENTS

Studies comparing LDLT and DDLT for HCC patients are summarized in Table 1. All DDLTs reviewed here were done with the whole liver graft.

Studies reporting a poorer outcome in the LDLT setting

Park *et al*[10] recently reported poorer recurrence-free survival among 166 LDLT recipients (81% at 5 years) compared to 50 DDLT recipients (94% at 5 years; $P = 0.045$). The noteworthy finding of this study was that the smaller the LDLT graft, the poorer the recurrence - free survival. Based on this finding, Park *et al*[10] suggested that the physiology of the small graft may stimulate tumor recurrence.

The results of the A2ALL cohort in United States also demonstrated an impaired outcome in LDLT recipients. In their initial report[11], they found a higher rate of recurrence within 3 years in LDLT than in DDLT (29% *vs* 0%, $P = 0.002$), but there was a clear tendency toward more aggressive tumor characteristics in the LDLT group. The same group recently published an updated report[12], in which HCC recurrence remained significantly different between LDLT and DDLT after adjustment for tumor characteristics. They concluded that the higher recurrence observed after LDLT was likely due to differences in the tumor characteristics, pretransplant HCC management, and waiting time.

Vakili *et al*[13] reporting the Lahey Clinic experience, demonstrated that the HCC recurrence rate of LDLT (29%) was significantly higher than that of DDLT (12%) ($P < 0.05$), but survival after LDLT was significantly better than that following DDLT for HCC during the same

period ($P = 0.02$).

Lo *et al*[14] from Hong Kong also reported a significantly higher incidence of HCC recurrence, 29% in LDLT and 0% in DDLT ($P = 0.029$). While the tumor characteristics were comparable between groups, the authors speculated that LDLT as a salvage transplantation, microscopic vascular invasion, and liver regeneration led to the difference in the recurrence rate.

Studies reporting a comparable outcome

Sandhu and colleagues of the Toronto group[15] reported that LDLT and DDLT both provide similarly low recurrence rates and high survival rates. They compared the results of 58 LDLT cases with those of 287 DDLT cases having comparable tumor characteristics, in which the 1-, 3-, and 5-year recurrence-free survival rates were 88%, 75%, and 70%, and 86%, 75%, and 70%, respectively.

In a well-designed study by Bhangui *et al*[16], an intention-to-treat analysis was conducted with recurrence rate representing the primary endpoint, comparing 36 LDLT cases and 147 DDLT cases. The authors demonstrated that both LDLT and DDLT provided similar recurrence - free survival rates (88% *vs* 86% at 5 years) for patients with HCC. The dropout rate and waiting time were significantly lower in the LDLT group than in the DDLT group, and there was also a trend toward a longer time to recurrence in the LDLT group, which may guarantee additional advantages with LDLT.

The Mount Sinai group[17,18] reported comparable recurrence - free survival between LDLT ($n = 36$) and DDLT ($n = 165$; 74% *vs* 83% at 2 years, $P = 0.3$). When stratified by tumor size (5 cm diameter) and the existence of microvascular invasion, there was still no difference between groups.

Sotiropoulos and colleagues of Essen, Germany[19,20], also supported the comparable recurrence - free survival rates between LDLT and DDLT for HCC (75% *vs* 81% at 3 years).

Hwang *et al*[21] of South Korea performed a nationwide survey regarding this issue. Among 237 LDLTs and 75 DDLTs for HCC, the 1 - and 3 - year recurrence - free survival rates were 83% and 80%, and 88% and 82%, respectively, with no significant difference between them.

A comparison of outcomes after liver transplantation obtained from database studies revealed comparable patient survival rates between LDLT and DDLT. According to a report from the Japanese Liver Transplantation Society Registry[22], a total of 6097 LDLTs were performed in Japan by the end of 2010, and 1225 (32%) were indicated for HCC, which was the most common indication in adult patients. The 1-, 3-, 5-, and 10-year cumulative survival rates of LDLT for HCC were 85%, 74%, 69%, and 60%, respectively. Todo and colleagues[23] performed a detailed survey using the same database (up to the end of 2005), comprising 653 patients who had undergone LDLT for HCC in Japan. At 1, 3, and 5 years, overall patient survival was 83%, 73%, and 69%, and disease-free survival was 77%, 65%, and 61%, respectively. Based on preoperative imaging studies, 62% were within the Milan criteria and 38% were beyond the Milan criteria, with 5-year recurrence-free survival rates of 90% and 61%, respectively ($P < 0.001$). These findings do not differ much from those obtained in the DDLT database of the United States and Europe[24-27], and may validate the use of LDLT for HCC patients.

CURRENT OPINIONS REGARDING THE DIFFERENCE BETWEEN LDLT AND DDLT

A randomized clinical study would be best to settle the controversy regarding the use of LDLT *vs* DDLT for HCC patients, but this is indeed difficult, if not impossible, to realize given the complicated decision-making process involved in LDLT. No prospective study has been conducted to date.

The Toronto group[28] recently performed a meta-analysis on 12 retrospective studies comparing the recurrence rates and recurrence - free survival between LDLT and DDLT recipients. A total of 633 LDLTs and 1232 DDLTs were enrolled, and the study provided evidence of lower disease - free survival after LDLT compared with DDLT for HCC (HR = 1.59, 95%CI: 1.02-2.49; $P = 0.041$). In contrast, there was no difference in overall survival between LDLT and DDLT (HR = 0.97, 95%CI: 0.73-1.27; $P = 0.808$). As mentioned by the authors of the paper, however, all involved studies were retrospective, had a low data quality score with poor reporting of baseline patient characteristics and an inadequate statistical approach, and were heterogeneous in critical aspects such as indication criteria and basal tumor characteristics, which warrant further well-designed studies to determine whether differences in HCC recurrence are due to study biases or biologic differences.

A recent review article by experts[29] concluded as follows: Although there is no strong evidence to support the higher HCC recurrence rates in LDLT than DDLT, the higher recurrence rates in LDLT recipients reported by several authors cannot be ignored. Actually, there are critical differences among societies such as: (1) differences in the allocation system for DDLT and LDLT; (2) differences in the availability of deceased donors; (3) differences in the potential waiting time; and (4) the differences in regional and national organ transplant law. In addition to taking into account these differences, liver transplant candidates with HCC and their potential live donors should be informed following risks and benefits; the waiting time for DDLT may lead to the dropout due to HCC progression which could be avoided by the prompt LDLT, however, the prompt LDLT may mask the aggressive tumor characteristics which may lead to a higher HCC recurrence rates. Although the currently available literatures can provide a low evidence for the difference of HCC recurrence between DDLT and LDLT, the tumor characteristics and biology seem to significantly influence on the recurrence, while the graft type and waiting time are less likely important as a possible risk factor.

Table 2 Graft characteristics and hepatocellular carcinoma recurrence

	Patients with recurrence (n = 11)	Patients without recurrence (n = 114)	P
Regeneration rate at 3 mo (%)	90 ± 24	93 ± 34	0.732
Graft type: right/left	4/7	36/78	0.702
Initial graft volume ratio to standard liver volume (%)	46 ± 9	47 ± 9	0.842

POSTULATED THEORIES FOR DIFFERENCES BETWEEN LDLT AND DDLT

LDLT provides several advantages compared with DDLT, such as a shorter waiting time, good quality graft with normal liver function and shorter ischemic time, and pretransplant treatment optimization, which might contribute to improved survival in LDLT recipients. Some of these characteristics, on the other hand, may lead to a favorable milieu for tumor progression[9].

There are several hypotheses other than tumor characteristics to explain the inferior outcome of LDLT. One explanation for the higher recurrence rates in LDLT is fast-tracking patients into liver transplantation, the so - called fast-track effect[11,30]. Some patients with more biologically aggressive HCC might drop off the waiting list due to tumor progression beyond the criteria during the wait-time in the DDLT setting. In contrast, due to the shortened wait time for LDLT candidates, progression of HCC with an aggressive tumor biology might not be recognized during such a short wait-time. This scenario might account for the higher HCC recurrence in the LDLT setting.

Another hypothesized mechanism for the higher recurrence rates in LDLT is that growth factors and cytokines released during rapid regeneration of the partial grafts from living donors might contribute to tumor progression and recurrence[31-34]. A rapidly regenerating liver parenchyma and ischemic-reperfusion injury facilitated by a small-for-size graft in LDLT setting might be a more favorable environment for tumor progression and HCC recurrence.

Additionally, some authors[11,35,36] insist that the technique of LDLT per se foregoes the principles of oncologic surgery. During LDLT, the meticulous dissection and mobilization of the liver might increase the possibility of tumor capsule violation or tumor embolization through the hepatic veins, thus promoting tumor dissemination. Preserving the native vena cava and the bile duct/hepatic artery/portal vein in the hepatic hilum might increase the risk of leaving the residual tumors.

As opposed with the above-mentioned anecdotal explanations, the advanced tumor characteristics of LDLT recipients can reasonably explain the higher recurrence rate in the LDLT setting. Grafts from living donors are not limited by restrictions imposed by the organ allocation system, meaning that the relation of the graft and recipient is usually one-on-one. Consequently, selection criteria based on the tumor burden, such as the tumor size and number, can be considered relative on a case-by-case basis, taking into account the presence of risk factors for recurrence and the chance of survival, as well as the wishes of the donor[37]. Consequently, the majority of Asian transplant centers have adopted extended criteria beyond those of Milan or the University of California, San Francisco (UCSF)[38]. Based on some studies, differences in patient tumor characteristics between LDLT and DDLT remain a main reason for the higher recurrence rate in LDLT. Additionally, in the majority of the aforementioned studies comparing LDLT and DDLT for HCC patients, tumor burdens such as the size, number, vascular invasion, and poor differentiation have proved to be independent risk factors for HCC recurrence after liver transplantation, all of which may lead to a rational explanation for the impaired recurrence - free survival of LDLT compared to DDLT.

OUR EXPERIENCE

At our institution, the University of Tokyo Hospital, a total of 423 adult recipients underwent LDLT by the end of 2012. Among them, 125 (30%) patients had HCC. The principle criterion for LDLT for HCC at our center is "up to 5 nodules with a maximum tumor diameter within 5 cm", which we call the "5-5 rule"[39]. Of the 125 patients, 118 (94%) were within the 5-5 rule criteria and 109 (87%) were within the Milan criteria. Overall survival of the 125 recipients at 1, 3, and 5 years was 88%, 82%, and 76%, respectively, with a median follow-up period of 8 years. A total of 11 (9%) patients developed HCC recurrence with a cumulative recurrence rate at 1, 3, and 5 years of 6%, 9%, and 11%, respectively.

We compared the graft regeneration rate between patients with HCC recurrence (n = 11) and those without recurrence (n = 114) to confirm the association of liver regeneration with HCC recurrence. The regeneration rate was calculated as follows: (graft volume at 3 mo after LDLT- initial graft volume)/initial graft volume × 100 (%). As shown in Table 2, there was no difference in the regeneration rate between those with HCC recurrence and those without recurrence. At the same time, the graft type (right *vs* left) and the initial graft volume ratio to the recipient's standard liver volume were also compared between groups, revealing no difference. A similar result was reported by the Asan group of South Korea[40], in which the graft-recipient weight ratio had no impact on HCC recurrence after LDLT among 181 LDLT recipients with HCC. Our result as well as the report of the Asan group clearly demonstrated that graft regeneration of the partial liver graft has no impact on HCC recurrence, at least in a clinical setting. The independent predictors for HCC recurrence in our series were tumors not within the 5-5 rule (Tokyo criteria), AFP level over 400 ng/mL, and des-

gamma-carboxy prothrombin levels over 200 mAU/mL.

CONCLUSION

In conclusion, there is no strong evidence to support higher HCC recurrence after LDLT than DDLT, and it may be reasonable to use different indication criteria for LDLT and DDLT, while there could be a potential bias in choosing the articles in the present study. LDLT should always be considered as a treatment option for HCC patients with advanced cirrhosis in areas where deceased donors are scarce or for patients whose tumor status interrupts access to DDLT.

REFERENCES

1 **Ferlay J**, Shin HR, Bray F, Forman D, Mathers C, Parkin DM. Estimates of worldwide burden of cancer in 2008: GLOBOCAN 2008. *Int J Cancer* 2010; **127**: 2893-2917 [PMID: 21351269 DOI: 10.1002/ijc.25516]

2 **Forner A**, Llovet JM, Bruix J. Hepatocellular carcinoma. *Lancet* 2012; **379**: 1245-1255 [PMID: 22353262 DOI: 10.1016/S0140-6736(11)61347-0]

3 **Mazzaferro V**, Regalia E, Doci R, Andreola S, Pulvirenti A, Bozzetti F, Montalto F, Ammatuna M, Morabito A, Gennari L. Liver transplantation for the treatment of small hepatocellular carcinomas in patients with cirrhosis. *N Engl J Med* 1996; **334**: 693-699 [PMID: 8594428 DOI: 10.1056/NEJM199603143341104]

4 **Bruix J**, Gores GJ, Mazzaferro V. Hepatocellular carcinoma: clinical frontiers and perspectives. *Gut* 2014; **63**: 844-855 [PMID: 24531850]

5 **Bruix J**, Sherman M. Management of hepatocellular carcinoma: an update. *Hepatology* 2011; **53**: 1020-1022 [PMID: 21374666 DOI: 10.1002/hep.24199]

6 **de Villa V**, Lo CM. Liver transplantation for hepatocellular carcinoma in Asia. *Oncologist* 2007; **12**: 1321-1331 [PMID: 18055852 DOI: 10.1634/theoncologist.12-11-1321]

7 **Lee Cheah Y**, K H Chow P. Liver transplantation for hepatocellular carcinoma: an appraisal of current controversies. *Liver Cancer* 2012; **1**: 183-189 [PMID: 24159583 DOI: 10.1159/000343832]

8 **Hwang S**, Lee SG, Belghiti J. Liver transplantation for HCC: its role: Eastern and Western perspectives. *J Hepatobiliary Pancreat Sci* 2010; **17**: 443-448 [PMID: 19885638 DOI: 10.1007/s00534-009-0241-0]

9 **Quintini C**, Hashimoto K, Uso TD, Miller C. Is there an advantage of living over deceased donation in liver transplantation? *Transpl Int* 2013; **26**: 11-19 [PMID: 22937787]

10 **Park MS**, Lee KW, Suh SW, You T, Choi Y, Kim H, Hong G, Yi NJ, Kwon CH, Joh JW, Lee SK, Suh KS. Living-donor liver transplantation associated with higher incidence of hepatocellular carcinoma recurrence than deceased-donor liver transplantation. *Transplantation* 2014; **97**: 71-77 [PMID: 24056623 DOI: 10.1097/TP.0b013e3182a68953]

11 **Fisher RA**, Kulik LM, Freise CE, Lok AS, Shearon TH, Brown RS, Ghobrial RM, Fair JH, Olthoff KM, Kam I, Berg CL. Hepatocellular carcinoma recurrence and death following living and deceased donor liver transplantation. *Am J Transplant* 2007; **7**: 1601-1608 [PMID: 17511683 DOI: 10.1111/j.1600-6143.2007.01802.x]

12 **Kulik LM**, Fisher RA, Rodrigo DR, Brown RS, Freise CE, Shaked A, Everhart JE, Everson GT, Hong JC, Hayashi PH, Berg CL, Lok AS. Outcomes of living and deceased donor liver transplant recipients with hepatocellular carcinoma: results of the A2ALL cohort. *Am J Transplant* 2012; **12**: 2997-3007 [PMID: 22994906 DOI: 10.1111/j.1600-6143.2012.04272.x]

13 **Vakili K**, Pomposelli JJ, Cheah YL, Akoad M, Lewis WD, Khettry U, Gordon F, Khwaja K, Jenkins R, Pomfret EA. Living donor liver transplantation for hepatocellular carcinoma: Increased recurrence but improved survival. *Liver Transpl* 2009; **15**: 1861-1866 [PMID: 19938113 DOI: 10.1002/lt.21940]

14 **Lo CM**, Fan ST, Liu CL, Chan SC, Ng IO, Wong J. Living donor versus deceased donor liver transplantation for early irresectable hepatocellular carcinoma. *Br J Surg* 2007; **94**: 78-86 [PMID: 17016793 DOI: 10.1002/bjs.5528]

15 **Sandhu L**, Sandroussi C, Guba M, Selzner M, Ghanekar A, Cattral MS, McGilvray ID, Levy G, Greig PD, Renner EL, Grant DR. Living donor liver transplantation versus deceased donor liver transplantation for hepatocellular carcinoma: comparable survival and recurrence. *Liver Transpl* 2012; **18**: 315-322 [PMID: 22140013 DOI: 10.1002/lt.22477]

16 **Bhangui P**, Vibert E, Majno P, Salloum C, Andreani P, Zocrato J, Ichai P, Saliba F, Adam R, Castaing D, Azoulay D. Intention-to-treat analysis of liver transplantation for hepatocellular carcinoma: living versus deceased donor transplantation. *Hepatology* 2011; **53**: 1570-1579 [PMID: 21520172 DOI: 10.1002/hep.24231]

17 **Gondolesi GE**, Roayaie S, Muñoz L, Kim-Schluger L, Schiano T, Fishbein TM, Emre S, Miller CM, Schwartz ME. Adult living donor liver transplantation for patients with hepatocellular carcinoma: extending UNOS priority criteria. *Ann Surg* 2004; **239**: 142-149 [PMID: 14745320 DOI: 10.1097/01.sla.0000109022.32391.eb]

18 **Roayaie S**, Schwartz JD, Sung MW, Emre SH, Miller CM, Gondolesi GE, Krieger NR, Schwartz ME. Recurrence of hepatocellular carcinoma after liver transplant: patterns and prognosis. *Liver Transpl* 2004; **10**: 534-540 [PMID: 15048797 DOI: 10.1002/lt.20128]

19 **Malagó M**, Sotiropoulos GC, Nadalin S, Valentin-Gamazo C, Paul A, Lang H, Radtke A, Saner F, Molmenti E, Beckebaum S, Gerken G, Frilling A, Broelsch CE. Living donor liver transplantation for hepatocellular carcinoma: a single-center preliminary report. *Liver Transpl* 2006; **12**: 934-940 [PMID: 16528715 DOI: 10.1002/lt.20677]

20 **Sotiropoulos GC**, Lang H, Nadalin S, Neuhäuser M, Molmenti EP, Baba HA, Paul A, Saner FH, Weber F, Hilgard P, Frilling A, Broelsch CE, Malagó M. Liver transplantation for hepatocellular carcinoma: University Hospital Essen experience and meta-analysis of prognostic factors. *J Am Coll Surg* 2007; **205**: 661-675 [PMID: 17964442 DOI: 10.1016/j.jamcollsurg.2007.05.023]

21 **Hwang S**, Lee SG, Joh JW, Suh KS, Kim DG. Liver transplantation for adult patients with hepatocellular carcinoma in Korea: comparison between cadaveric donor and living donor liver transplantations. *Liver Transpl* 2005; **11**: 1265-1272 [PMID: 16184545 DOI: 10.1002/lt.20549]

22 Liver transplantation in Japan- registry by the Japnese Liver Transplantation Society. *Ishoku* 2012; **46**: 524-536

23 **Todo S**, Furukawa H, Tada M. Extending indication: role of living donor liver transplantation for hepatocellular carcinoma. *Liver Transpl* 2007; **13**: S48-S54 [PMID: 17969069]

24 **Mazzaferro V**, Bhoori S, Sposito C, Bongini M, Langer M, Miceli R, Mariani L. Milan criteria in liver transplantation for hepatocellular carcinoma: an evidence-based analysis of 15 years of experience. *Liver Transpl* 2011; **17** Suppl 2: S44-S57 [PMID: 21695773]

25 **Adam R**, Karam V, Delvart V, O'Grady J, Mirza D, Klempnauer J, Castaing D, Neuhaus P, Jamieson N, Salizzoni M, Pollard S, Lerut J, Paul A, Garcia-Valdecasas JC, Rodríguez FS, Burroughs A. Evolution of indications and results of liver transplantation in Europe. A report from the European Liver Transplant Registry (ELTR). *J Hepatol* 2012; **57**: 675-688 [PMID: 22609307 DOI: 10.1016/j.jhep.2012.04.015]

26 **Singal AK**, Guturu P, Hmoud B, Kuo YF, Salameh H, Wiesner RH. Evolving frequency and outcomes of liver transplantation based on etiology of liver disease. *Trans-*

plantation 2013; **95**: 755-760 [PMID: 23370710 DOI: 10.1097/TP.0b013e31827afb3a]
27 **Taniguchi M**. Liver transplantation in the MELD era--analysis of the OPTN/UNOS registry. *Clin Transpl* 2012; **2012**: 41-65 [PMID: 23721009]
28 **Grant RC**, Sandhu L, Dixon PR, Greig PD, Grant DR, McGilvray ID. Living vs. deceased donor liver transplantation for hepatocellular carcinoma: a systematic review and meta-analysis. *Clin Transplant* 2013; **27**: 140-147 [PMID: 23157398 DOI: 10.1111/ctr.12031]
29 **Grant D**, Fisher RA, Abecassis M, McCaughan G, Wright L, Fan ST. Should the liver transplant criteria for hepatocellular carcinoma be different for deceased donation and living donation? *Liver Transpl* 2011; **17** Suppl 2: S133-S138 [PMID: 21634006]
30 **Kulik L**, Abecassis M. Living donor liver transplantation for hepatocellular carcinoma. *Gastroenterology* 2004; **127**: S277-S282 [PMID: 15508095]
31 **Man K**, Lo CM, Xiao JW, Ng KT, Sun BS, Ng IO, Cheng Q, Sun CK, Fan ST. The significance of acute phase small-for-size graft injury on tumor growth and invasiveness after liver transplantation. *Ann Surg* 2008; **247**: 1049-1057 [PMID: 18520234 DOI: 10.1097/SLA.0b013e31816ffab6XXX]
32 **Shi JH**, Huitfeldt HS, Suo ZH, Line PD. Growth of hepatocellular carcinoma in the regenerating liver. *Liver Transpl* 2011; **17**: 866-874 [PMID: 21542129 DOI: 10.1002/lt.22325]
33 **Efimova EA**, Glanemann M, Liu L, Schumacher G, Settmacher U, Jonas S, Langrehr JM, Neuhaus P, Nüssler AK. Effects of human hepatocyte growth factor on the proliferation of human hepatocytes and hepatocellular carcinoma cell lines. *Eur Surg Res* 2004; **36**: 300-307 [PMID: 15359093 DOI: 10.1159/000079915]
34 **Man K**, Fan ST, Lo CM, Liu CL, Fung PC, Liang TB, Lee TK, Tsui SH, Ng IO, Zhang ZW, Wong J. Graft injury in relation to graft size in right lobe live donor liver transplantation: a study of hepatic sinusoidal injury in correlation with portal hemodynamics and intragraft gene expression. *Ann Surg* 2003; **237**: 256-264 [PMID: 12560784 DOI: 10.1097/01.SLA.0000048976.11824.67]
35 **Di Sandro S**, Slim AO, Giacomoni A, Lauterio A, Mangoni I, Aseni P, Pirotta V, Aldumour A, Mihaylov P, De Carlis L. Living donor liver transplantation for hepatocellular carcinoma: long-term results compared with deceased donor liver transplantation. *Transplant Proc* 2009; **41**: 1283-1285 [PMID: 19460539 DOI: 10.1016/j.transproceed.2009.03.022]
36 **Li C**, Wen TF, Yan LN, Li B, Yang JY, Wang WT, Xu MQ, Wei YG. Outcome of hepatocellular carcinoma treated by liver transplantation: comparison of living donor and deceased donor transplantation. *Hepatobiliary Pancreat Dis Int* 2010; **9**: 366-369 [PMID: 20688599]
37 **Tamura S**, Sugawara Y, Kokudo N. Living donor liver transplantation for hepatocellular carcinoma: the Japanese experience. *Oncology* 2011; **81** Suppl 1: 111-115 [PMID: 22212944]
38 **Chan SC**. Liver transplantation for hepatocellular carcinoma. *Liver Cancer* 2013; **2**: 338-344 [PMID: 24400221 DOI: 10.1159/000343849]
39 **Sugawara Y**, Tamura S, Makuuchi M. Living donor liver transplantation for hepatocellular carcinoma: Tokyo University series. *Dig Dis* 2007; **25**: 310-312 [PMID: 17960065 DOI: 10.1159/000106910]
40 **Hwang S**, Lee SG, Ahn CS, Kim KH, Moon DB, Ha TY, Park KM, Song GW, Jung DH, Kim BS, Moon KM. Small-sized liver graft does not increase the risk of hepatocellular carcinoma recurrence after living donor liver transplantation. *Transplant Proc* 2007; **39**: 1526-1529 [PMID: 17580180 DOI: 10.1016/j.transproceed.2007.03.066]

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Impact of Donor and Recipient Single Nucleotide Polymorphisms of IL28B rs8099917 in Living Donor Liver Transplantation for Hepatitis C

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Abstract

Single nucleotide polymorphisms of interleukin-28B (IL28B) rs8099917 are reported to be associated with virologic clearance in interferon-and ribavirin -based treatment for hepatitis C virus (HCV)-infected patients. We examined virologic response in accordance with IL28B polymorphisms in our living donor liver transplantation series under a preemptive interferon and RBV treatment approach. Adequate DNA samples from both the recipient and donor for the study of single nucleotide polymorphisms of IL28B were available from 96 cases and were the subjects of the present study. Various clinical factors related with virologic response including early virologic response (EVR) and sustained virologic response (SVR) were examined. Totally 51% presented with EVR and 44% achieved SVR. Presence of the major allele (TT) in either the recipient or the donor corresponded to SVR of 53% and 48%. Presence of the minor allele (TG or GG) corresponded to SVR of 26% and 32%. Multivariate analysis revealed that genotype of HCV or EVR, but not IL28B polymorphisms in either the recipient or donor, was an independent factor for achieving SVR. When virologic response to treatment was incorporated into analysis, the impact of IL28B polymorphism on virological clearance remained relative to other factors and was not significantly independent.

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Introduction

Hepatitis C virus (HCV) infection is the leading cause of end-stage liver disease necessitating liver transplantation in developed countries [1–3]. HCV reinfection following liver transplantation is universal, however, and the histologic progression of HCV-related liver cirrhosis is accelerated in comparison with the non-transplant population [4–8]. Long-term outcomes are reported to be poorer in liver transplant recipients with HCV recurrence [9–10]. Although its efficacy remains unsatisfactory, the standard treatment for HCV relapse following liver transplantation is the combined application of pegylated interferon and ribavirin (RBV) [11–16]. To improve the overall outcomes of patients undergoing living donor liver transplantation (LDLT) for HCV-related liver disease, we routinely administered interferon based treatment preemptively in our series [17] with recent results indicating a sustained viral response (SVR) rate of 43% [18].

In a recent genome-wide association study, three independent institutions identified single nucleotide polymorphisms in the interleukin (IL)-28B gene on chromosome 19q13; rs12980275 or rs8099917 as being strongly associated with the virologic response to interferon and RBV-based treatment in HCV-infected patients [19–21]. In Japanese patients, the G nucleotide of rs8099917 (minor allele) is associated with a poor response to treatment, whereas a T nucleotide (major allele) is associated with a fair response [20].

The impact of the IL28B polymorphism was recently studied in the liver transplant setting. A small series reported that combined analysis of IL28B polymorphisms in both donors and recipients may predict the possibility of achieving SVR under current standard pegylated interferon-and RBV-based therapy [22–23]. The evidence, however, remains limited. In the present study, we examined the virologic response to a preemptive treatment approach in accordance with IL28B polymorphism.

Patients and Methods

Patients

From June 1996 to December 2012, 499 LDLT surgeries were performed at the University of Tokyo Hospital among which 134 cases were for HCV-related liver disease. Among them, three recipients remained negative for HCV RNA after transplantation

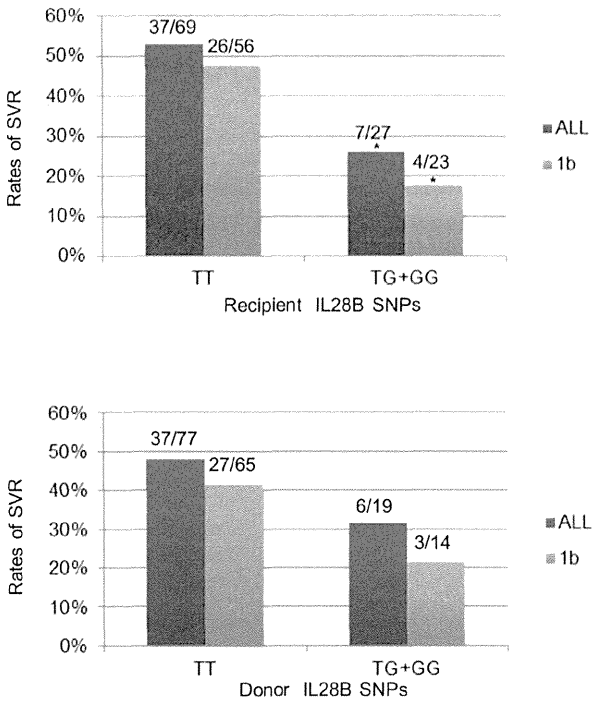

Figure 1. IL28B rs8099917 SNPs in the recipient and donor and corresponding rates of SVR. * indicates p = <0.05 v.s. major allele (TT) with corresponding background.
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alone, not requiring interferon treatment, and were excluded from the study. In the remaining 131, either appropriate informed consent or adequate samples for genetic analysis was not available in 25 cases for the following reasons and therefore excluded; death during the earlier period in 18, lost to follow-up in 3, and decline of study consent in 14 cases. The remaining 96 cases were the subjects of the present analysis. The earliest case of this population underwent LDLT in June 1998, and the latest case underwent LDLT in February 2011. Follow up was done until the end of 2012, or death.

Antiviral therapy for HCV

Treatment was initiated with low-dose interferon alpha-2b and RBV 400 mg/day promptly after improvement of the general condition following liver transplantation. Recovery of hematologic and renal function was considered crucial. Specifically, initiation was considered when the leukocyte number > = 4000/ml, platelet count > = 50 000/ml, hemoglobin > = 8 g/l, and serum creatinine < 2 mg/dl. Thereafter, the dosage was gradually increased as tolerated. Finally, pegylated-interferon alpha-2b 1.5 μg/kg/week and RBV 800 mg/day were administered, depending on patient compliance [17,18].

HCV RNA was measured quantitatively by reverse-transcriptase polymerase chain reaction (Amplicor HCV; Roche Molecular Systems, Pleasanton, CA). The response was considered to be an EVR at 12 week with an at least 2 log10 drop in serum HCV-RNA. The treatment was continued for 12 months after serum HCV-RNA turned negative, defined as ETR. The response was considered to be a SVR after another 6 months of negative serologic results without anti-viral treatment. Flexible dose adjustments were made as necessary to avoid serious adverse events and to prevent a lapse in treatment. A fixed overall treatment period length was not defined [17,18].

Analysis of the genotype of IL28B

Genomic DNA from recipients and donors was extracted either from peripheral blood mononuclear cells or from formalin-fixed paraffin-embedded liver biopsy samples depending on availability. Genotyping of single nucleotide polymorphisms IL28B rs8099917 was performed as previously described [20,22].

Ethics Statement

All LDLTs were performed after individually obtaining informed consent from recipients and donors. LDLT program at the University Of Tokyo Hospital has been approved by it's Institutional Review Board, and all aspects of the procedures have been conducted according to the principles expressed in the Declaration of Helsinki. The current human subject research was approved as project number G3514 by Graduate School of Medicine and Faculty of Medicine, the University of Tokyo Research Ethics Committee and Human Genome, Gene Analysis Research Ethics Committee. All subjects have been properly instructed and participated by signing the appropriate informed consent paperwork. In the preparation of this manuscript, all efforts have been made to protect patient privacy and anonymity.

Statistical analysis

The cumulative rate of viral responses was studied using the Kaplan-Meier method. Comparison was made using the log-rank test. A multivariate analysis was performed using the Cox proportional-hazards model and a forward stepwise procedure. Categorical various clinical factors were compared between groups using the Fisher's exact test. JMP 9 software (SAS Institute Japan, Tokyo, Japan) was used for all analyses. A p-value of less than 0.05 was considered statistically significant.

Results

Patient demographics and virologic response

All recipients and donors of the 96 cases were of Japanese origin. Median age of the recipients and donors was 56 (range 23–66) and 34 (range 17–66), respectively. In 60 cases, the donor was a relative within a first degree of consanguinity. Among the recipients, 60 cases presented with hepatocellular carcinoma, 3 with HIV co-infection, and 2 with hepatitis B virus co-infection. The HCV RNA genotype was confirmed to be 1b in 79 (81%). Median calculated model for end-stage liver disease score at the time of transplant was 14.

All 96 recipients received interferon treatment according to our pre-emptive regimen described above. None presented with fibrosing cholestatic hepatitis at the time of treatment initiation. Overall, at the time of follow-up, 49 (51%) presented with early virologic response (EVR). Sixty-five (67%) patients had negative serum HCV RNA results at least once, among which 52 (54%) patients experienced a non-detectable level of serum HCV RNA for 12 months on treatment (end of treatment response, ETR). Forty-three (44%) patients remained negative for serum HCV RNA for 6 months or longer after cessation of interferon treatment following ETR, and these recipients were considered to have achieved SVR. Consistent with the nature of a treatment protocol without a defined time endpoint, the response rate increased over time. The cumulative rates of SVR and ETR based on the Kaplan-Meier method are 41% and 54%, respectively, 5 years after transplantation.

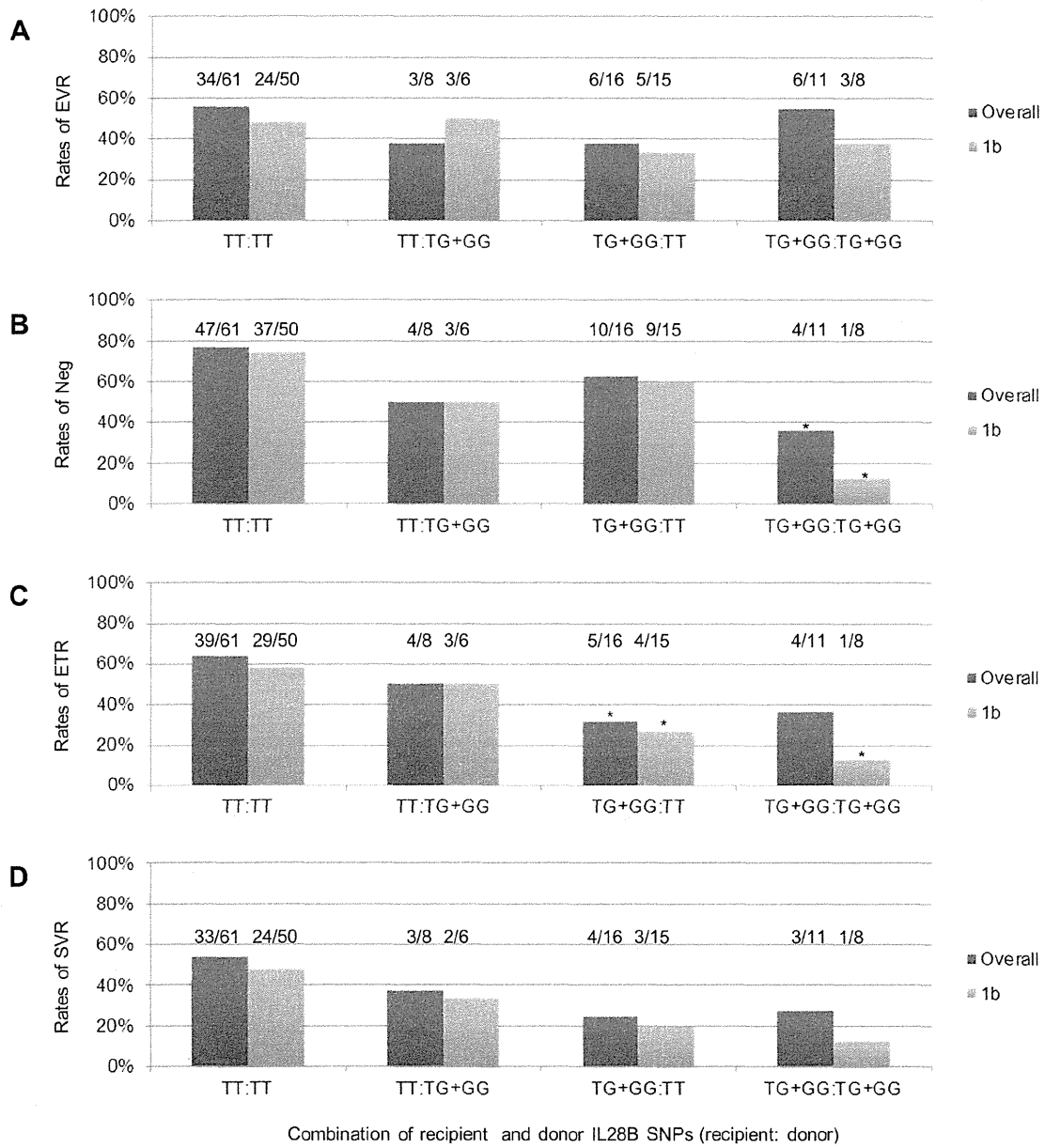

Figure 2. Combination of recipient and donor IL28B rs8099917 SNPs and corresponding rates of viral responses (A–D). A. Rates of EVR, B. Rates of recipients that presented with temporal non-detectable level of serum HCV RNA at least once during the course of INF treatment, C. rates of ETR, D. rates of SVR. Abbreviation: SNPs, single nucleotide polymorphism; SVR, sustained viral response; EVR, early virologic response; Neg, temporal non-detectable level of serum HCV RNA; ETR, end of treatment response. * indicates p = <0.05 v.s. combination of major allele both in the recipient and donor (TT:TT) with corresponding background.
doi:10.1371/journal.pone.0090462.g002

Frequency of IL28B rs8099917 single nucleotide polymorphisms in recipients and donors

The proportion of TT, TG and GG genotypes in recipients was 72%, 28%, and 0%, respectively, whereas TT was the most frequent genotype in donors (80%), followed by genotypes TG (19%) and GG (1%). This distribution did not differ significantly between genotype 1b and the others.

IL28B polymorphism and interferon sensitivity after LDLT

As previously reported, IL28B polymorphisms in both the recipient and donor seemed to be strongly correlated with the sensitivity to interferon treatment. The major allele (TT) in the recipient and donor corresponded to SVR rates of 54% (37 of 69) and 48% (37 of 77), respectively, whereas the presence of the minor allele (TG or GG) corresponded to SVR rates of 26% (7 of 27) and 32% (6 of 19), respectively (Figure 1). Difference of SVR between TT and TG/GG in recipient was statistically significant

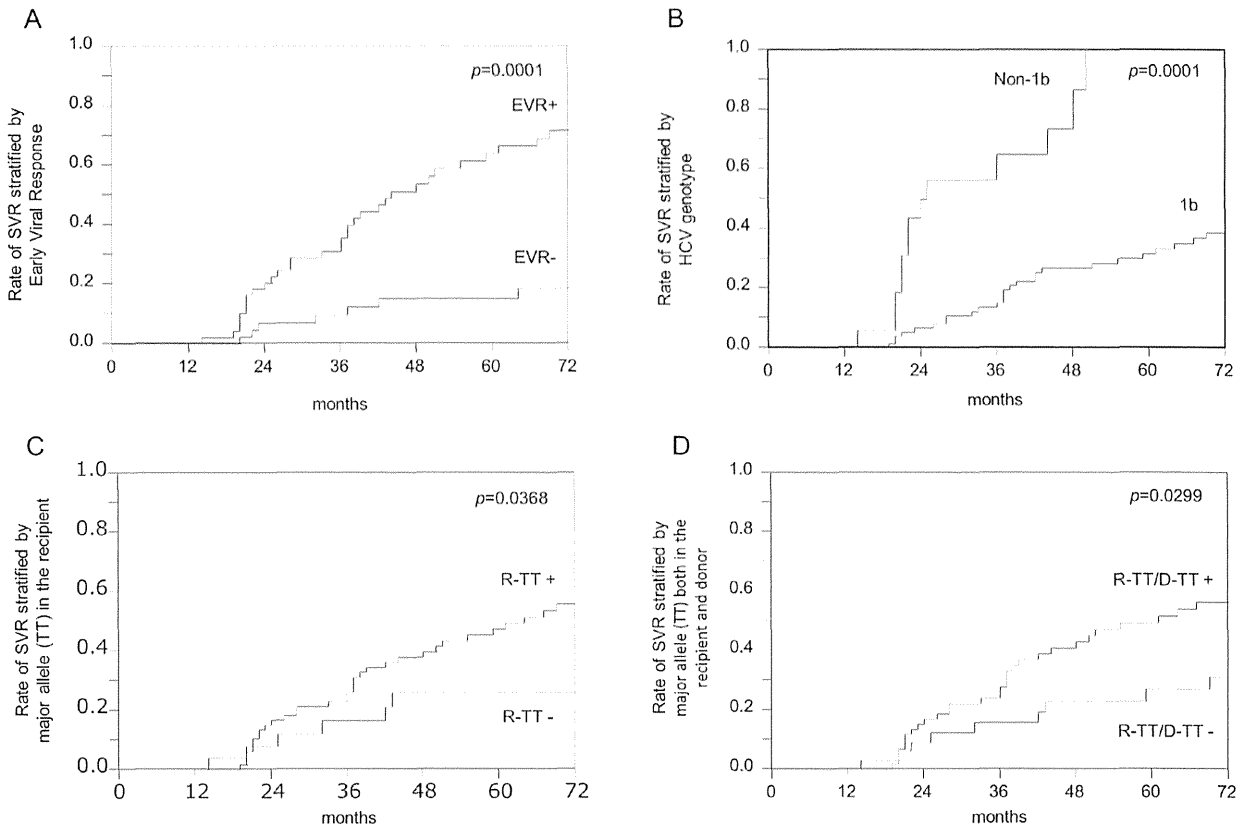

Figure 3. Cumulative rates of SVR based on the Kaplan-Meier method. A. Stratified by EVR, B. HCV genotype, C. major allele (TT) in the recipient, and D. major allele (TT) both in the recipient and donor. Abbreviations: SVR, sustained viral response; EVR, Early viral response; R-TT, major allele (TT) in the recipient; D-TT, major allele (TT) in the donor.
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($p = 0.0219$ and $p = 0.0212$ for all recipients and for recipients with genotype 1b HCV, respectively). However, difference of SVR according to the donor IL28B SNPs did not reach statistical significance ($p = 0.3029$ and $p = 0.2279$ for all donors and for donors whose recipients with genotype 1b, respectively).

The combinations of recipient and donor IL28B polymorphisms and corresponding rates of virologic responses are summarized in Figures 2A–2D. Although combinations of recipients and grafts obtained from donors both carrying the major homozygous allele presented with tendency of higher rates of virologic eradication demonstrated as ETR (Figure 2C) or SVR (Figure 2D), especially in the case of HCV RNA genotype 1b, this synergistic tendency remained unclear or limited when observation was extended to on-treatment virologic response at an earlier stage evaluated by temporal clearance or EVR (Figures 2A and 2B). For example, rates of EVR between recipient and donor pairs carrying both a major homozygous allele and minor allele did not differ significantly ($p = 0.9416$), and the advantage remained unclear even when limited to the HCV RNA genotype 1b ($p = 0.5804$) (Figure 2A). Similarly, the presence of a minor allele either in the recipient or the donor did not significantly affect temporal viral clearance (Figure 2B).

Impact of IL28B polymorphism among other factors

Impact of various clinical factors on sensitivity to interferon treatment in the present study was assessed by SVR rates (Table 1). Compatible with previous studies, univariate analysis revealed that HCV genotype 1b and presence of EVR were significant factors affecting outcome. As for IL28B polymorphisms, a major allele homozygote in the recipient, and a major allele homozygote both in the recipient and donor presented with a statistically significant impact ($p = 0.0368$, and 0.0299, respectively). A major allele homozygote in the donor side alone did not have strong impact (0.3136; Table 1). The above four factors significantly impacted SVR in univariate analysis are summarized in Figure 3.

To elucidate the magnitude of the IL28B polymorphism, a multivariate study was conducted including all clinical variables from Table 1. To incorporate the nature of our treatment protocol without a defined period of interferon treatment, multivariate analysis was performed using the Cox proportional-hazards model. The study revealed the genotype of HCV and EVR, but not the IL28B polymorphism of either recipient or donor, to be independent factors to achieve SVR. Recipients re-infected with HCV genotype 1b presented with a significantly poorer chance of SVR (Hazard ratio 0.277, 95% confidence interval 0.132–582, $p = 0.0007$). On the other hand, once EVR was observed, recipients demonstrated significantly better opportunity for SVR (Hazard ratio 4.426, 95% confidence interval 1.958–10.007, $p = 0.0004$). Cumulative rates of SVR within the genotype 1b or non-1b population stratified by the presence of EVR are presented in Figure 4. Conversely, background factors of recipients presenting EVR or not so depending on HCV RNA genotypes

Table 1. Sustained viral response in patients with combined treatment and clinical factors.

Factors		No.	%SVR at 5 y	p
R-age	=<60	72	48	
	>60	24	26	0.5221
R-sex	Male	70	44	
	Female	26	34	0.5895
MELD	=<15	52	42	
	>15	44	40	0.9585
HIV	Positive	3	67	
	Negative	93	40	0.0729
HCC	Positive	60	41	
	Negative	36	48	0.7344
Genotype	1b	79	32	
	Non-1b	17	100	<0.0001
Graft size	<50%	53	46	
%R-SLV	>=50%	43	37	0.8251
HCV-RNA	=<5.6	48	45	
titer	>5.6	48	38	0.2999
EVR	Yes	49	64	
	No	47	15	<0.0001
ACR	Yes	20	49	
	No	76	39	0.3844
D-age	=<40	61	39	
	>40	35	46	0.1101
D-sex	Male	62	33	
	Female	34	59	0.1155
CyA	Yes	58	51	
	No	38	27	0.0683
R-IL28B	TT	69	47	
	TG/GG	27	26	0.0368
D-IL28B	TT	77	44	
	TG/GG	19	31	0.3136
R-TT/D-TT	Yes	61	49	
	No	35	27	0.0299
R-TT/D-TG+GG	Yes	8	31	
	No	88	42	0.7276
R-TG+GG/D-TT	Yes	16	22	
	No	80	45	0.0962
R-TG+GG/D-TG+GG	Yes	11	31	
	No	85	43	0.3386

Abbreviations: No., number of patients; %SVR, rate of recipients achieving sustained viral response; R-age, age of the recipient at the time of transplantation; R-sex, sex of the recipient; MELD, Model for end-stage liver disease score; HIV, human immune deficiency virus; HCC, hepatocellular carcinoma; HCV, hepatitis C virus; %R-SLV, percentage of graft size to recipient's standard liver volume; HCV-RNA, hepatitis C viral ribonucleic acid; EVR, early viral response; ACR, acute cellular rejection; D-age, age of the donor at the time of transplantation; D-sex, sex of the donor; CyA, cyclosporine A; R-IL28B, recipient's IL28B polymorphism (rs8099917); D-IL28B, donor's IL28B polymorphism (rs8099917); R-TT/D-TT, Both recipient and donor carrying major allele (TT); R-TT/D-TG+GG, recipient carrying major allele (TT) but donor carrying minor allele (TG or GG); R-TG+GG/D-TT, recipient carrying minor allele (TG or GG) but donor carrying major allele (TT); R-TG+GG/D-TG+GG, Both recipient and donor carrying minor allele (TG or GG).

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were evaluated (Table 2). Among various clinical factors, IL28B polymorphisms, either that of the recipient or the donor, or both, was not prevalent in relation to EVR, especially in the genotype 1b population.

Overall, IL28B polymorphisms had a relative, not independent, impact. On-treatment virologic response to interferon and RBV-based treatment represented by EVR remain the most significant factor predicting SVR, even among recipients with HCV genotype 1b.

Discussion

Treatment of HCV re-infection by interferon and ribavirin after liver transplantation has remained a challenge with inferior outcomes compared to the non-transplantation population due to immunosuppression, and low tolerability. Recurrence and persistence of HCV-infection remain the most common cause of post-transplant graft loss and mortality. Identifying factors affecting the outcomes, including the response to treatment, therefore, continues to be a subject of keen interest. This study presents observations that may be potentially important in light of advancements involving recent genetic discoveries regarding IL28B polymorphisms.

This is the largest study to date, 96 LDLT cases, evaluating the impact of IL28B polymorphisms in both donor and recipient, in accordance with the on-treatment response, on the outcome. Also, the series is the first to analyze the magnitude of the polymorphism under a preemptive treatment approach [17,18] after LDLT. The study is limited to rs8099917 based on previous studies of IL28B SNPs in the Japanese population, and therefore, impact of rs12979860 awaits further similar study in the West.

Several predictive factors for interferon and RBV sensitivity in the non-transplant population were recently identified. Virologically, HCV-genotype, and HCV RNA mutations in the core and NS5A regions are recognized as important factors [24,25]. As for host factors, by genome-wide association study coming from three independent studies, IL28B polymorphisms have been identified as significant factors affecting virologic clearance [19–21]. The mechanism underlying the influence of IL28B polymorphisms on the response to interferon and RBV therapy is, however, yet to be determined. Current understanding is that the product of the IL28B gene is interferon lambda-3, which belongs to the type III interferon family that induces interferon-stimulated genes. Favorable IL28B polymorphisms are associated with decreased levels of intrahepatic interferon-stimulated genes, offering a favorable environment for virologic clearance under interferon and RBV treatment [26–28]. Whether or not this mechanism is applicable to the liver transplant setting, in which a liver allograft is infected by HCV under immunosuppression, remains to be studied. There is little evidence to speculate otherwise at this point.

Fukuhara and colleagues [22] first reported the impact of IL28B polymorphisms on the outcome of LDLT. In their study, IL28B polymorphisms were studied in 67 HCV infected recipients and 41 living liver donors. Interestingly, they reported that SVR achievement was significantly associated with IL28B polymorphisms of both the recipient and donor. When both were major-allele homozygotes, the SVR rate was 56%. Whereas when either the recipient or the donor presented with a minor heterozygote or homozygote allele, SVR rate dropped to 10%, and further, when both the recipient and donor presented with a minor heterozygote or homozygote allele, none achieved SVR. Kawaoka and colleagues [23] conducted a similar study involving 20 LDLT recipient and donor pairs. They reported that major-allele homozygotes in both the recipient and donor resulted in an

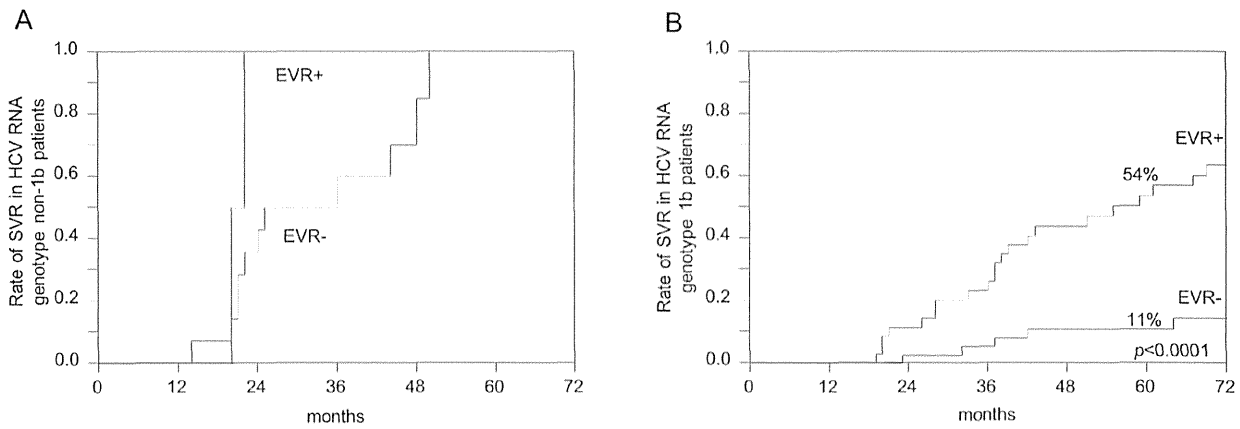

Figure 4. Cumulative rates of SVR stratified by EVR using the Kaplan-Meier method. A. HCV RNA genotype non-1b, B. HCV RNA genotype 1b. Abbreviations: SVR, sustained viral response; EVR, Early viral response.
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SVR rate of 81%. Although the number of cases where rather small, multivariate analysis including adherence to RBV therapy was performed, revealing that major-allele homozygotes in both the recipient and donor as the only independent and dominant determinant of SVR with and an odds ratio of 15.

Although logistic and technical difficulties remain in sampling and analyzing IL28B gene of the donors, comparable studies have been performed in the deceased donor setting [29–34]. The impact of IL28B polymorphisms has also become recognized in deceased donor liver transplantation for HCV. It was suggested that, patients requiring liver transplantation due to end-stage chronic HCV appeared to be selected toward the adverse genotypes [31], and the polymorphism seems to influence the degree of graft inflammation at biochemical and histologic levels following transplantation [29,32]. It has also become evident that, while there seems to be little doubt that IL28B polymorphisms markedly affect the response to interferon and RBV treatment, whether the donor or recipient, or the combination of both, should be considered paramount differ among studies. Lange and colleagues provide evidence that the donor's rather than the recipient's IL28B genetic background has a dominant impact on the virologic response [31], while Cotpo-Llerena et al. [34] report that the recipient's genetic background plays a major role. On the other hand, a recent study by Duarte-Rojo and colleagues [30] used multivariate analysis to demonstrate that the combination of both is the most influential. A German study [32] provided no data on the potential role of donor IL28B polymorphisms. The numbers of subjects in these studies remain small in comparison with the size of patients involved in analysis of non-transplant cases. Data on previously reported important clinical factors other than IL28B polymorphisms are not readily available for evaluation; much less an analysis by multivariate analysis to weigh the impact in a more reliable context. Clearly, further studies are required with an inclusion of a broader range of clinical data.

In our study, we included factors previously reported to influence the outcomes of the virologic response against interferon and RBV therapy in the analysis. This includes age and sex of both the donor and recipient, preoperative viral load, immunosuppression, and other factors as well as the on-treatment results represented by EVR (Table 1). A recent report by Thompson and colleagues suggests that on-treatment virologic response may have strong predictive power regardless of the IL28B type [35]. We used multivariate analysis that included all of these factors, and found that IL28B polymorphism is an influential but not determinant factor for SVR. Rather, in our series with a preemptive treatment approach, we demonstrated that on-treatment response was the key factor for predicting SVR.

Our study has three major weaknesses. First, although clinical virologic response was followed up and recorded in a prospective manner, IL28B polymorphisms were recently determined, making our study a retrospective case series with diverse sources of DNA. DNA samples for analysis were collected either from peripheral blood mononuclear cells or from formalin-fixed paraffin-embedded samples based on availability. A prospective study with a fixed DNA sampling protocol is required. Second, the nucleotide sequences of the core and non-structural 5A regions, another recently suggested important factor [36], have not been investigated in concert with IL28B polymorphisms. In fact, few studies to date have performed a combined analysis of both IL28B polymorphisms and HCV RNA nucleotide sequences, most likely due to the additional logistic burden. Fukuhara and colleagues [22] reported the synergistic value of combining findings from IL28B polymorphisms and HCV RNA nucleotide sequences in predicting the treatment response. This aspect should also be considered in future studies. Third, the study lacks histologic data. In our series, protocol biopsy was not performed. Hepatic venous gradient to evaluate the degree of liver fibrosis was also not routinely performed. This is due in part to our preemptive treatment strategy. Eurich and colleagues [32] have presented interesting outcomes regarding the progression of the histologic response in their deceased donor series. Comparable analysis in the living donor setting in the future may be valuable.

Finally, in the current study, direct antiviral agents in combination with peg-IFN and ribavirin were not used. Although the efficacy of the earlier generation of direct antiviral agents has become recognized in the non-transplant population, drastically altering standard treatment [37–39], its safety and effectiveness under routine use in the transplant population await future confirmation. Development in this aspect, however, is in rapid progression. Current recognition is that new-age anti-HCV treatment incorporating advanced direct antiviral agents will radically alter the outcome [40]. Further accumulation of data in combination with IL28B and the development of additional treatment options may be beneficial.

Table 2. Early viral response in patients with combined treatment and clinical factors.

		HCV RNA Genotype					
		Non-1b			1b		
Factors		EVR		p	EVR		p
		No	Yes		No	Yes	
R-age	=<60	2	11		31	28	
	>60	1	3	1.0000	13	7	0.4368
R-sex	Male	2	10		33	25	
	Female	1	4	1.0000	11	10	0.8001
MELD	=<15	3	5		26	18	
	>15	0	9	0.0824	18	17	0.6488
HIV	Positive	0	3		0	0	
	Negative	3	11	1.0000	44	35	NA
HCC	Positive	2	5		28	25	
	Negative	1	9	0.5368	16	10	0.4826
Graft size	<50%	1	7		26	19	
%R-SLV	>=50%	2	7	1.0000	18	16	0.8194
HCV-RNA titer	=<5.6	3	8		24	13	
	>5.6	0	6	0.5147	20	22	0.1735
ACR	Yes	0	2		7	11	
	No	3	12	1.0000	37	24	0.1149
D-age	=<40	1	7		28	25	
	>40	2	7	1.0000	16	10	0.4826
D-sex	Male	2	8		27	25	
	Female	1	6	0.6704	17	10	0.4744
CyA	Yes	0	1		19	18	
	No	3	13	1.0000	25	17	0.5029
R-IL28B	TT	3	10		29	27	
	TG/GG	0	4	0.5412	15	8	0.3251
D-IL28B	TT	1	11		36	29	
	TG/GG	2	3	0.1912	8	6	1.0000
R-TT/D-TT	Yes	1	10		26	24	
	No	2	4	0.5147	18	11	0.4826
R-TT/D-TG+GG	Yes	2	0		3	3	
	No	1	14	0.0221	41	32	1.0000
R-TG+GG/D-TT	Yes	0	1		10	5	
	No	3	13	1.0000	34	30	0.3983
R-TG+GG/D-TG+GG	Yes	0	3		5	3	
	No	3	11	1.0000	39	32	1.0000

Abbreviations: HCV-RNA, hepatitis C viral ribonucleic acid; EVR, early viral response; R-age, age of the recipient at the time of transplantation; R-sex, sex of the recipient; MELD, Model for end-stage liver disease score; HIV, human immune deficiency virus; HCC, hepatocellular carcinoma; HCV, hepatitis C virus; %R-SLV, percentage of graft size to recipient's standard liver volume; ACR, acute cellular rejection; D-age, age of the donor at the time of transplantation; D-sex, sex of the donor; CyA, cyclosporine A; R-IL28B, recipient's IL28B polymorphism (rs8099917); D-IL28B, donor's IL28B polymorphism (rs8099917); R-TT/D-TT, Both recipient and donor carrying major allele (TT); R-TT/D-TG+GG, recipient carrying major allele (TT) but donor carrying minor allele (TG or GG); R-TG+GG/D-TT, recipient carrying minor allele (TG or GG) but donor carrying major allele (TT); R-TG+GG/D-TG+GG, Both recipient and donor carrying minor allele (TG or GG).

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Conclusions

In contrast to previous reports, when virologic response to treatment was incorporated into analysis, the impact of IL28B polymorphism on achieving SVR remained relative in our living donor liver transplantation series under a preemptive interferon and RBV-based treatment approach. HCV genotype 1b and on-treatment response represented by EVR were both significant and independent factors. Caution should be used when incorporating the IL28B polymorphism into the treatment strategy of HCV reinfection following liver transplantation in an absolute manner, such as to the donor selection or graft allocation, however, until the mechanism of its effect is elucidated and well-designed future studies have confirmed its true nature.

Author Contributions

Conceived and designed the experiments: NH ST Y. Sugawara. Performed the experiments: JT TI JK TA Y. Sakamoto. Analyzed the data: KH TT NY. Contributed reagents/materials/analysis tools: NK. Wrote the paper: NH ST Y. Sugawara.

References

1. Lauer GM, Walker BD. (2001) Hepatitis C virus infection. N Engl J Med 345: 41–52.
2. Zeuzem S, Berg T, Moeller B, Hinrichsen H, Mauss S, et al. (2009) Expert opinion on the treatment of patients with chronic hepatitis C. J Viral Hepat. 16: 75–90.
3. Brown RS. (2005) Hepatitis C and liver transplantation. Nature 436:973–978.
4. Berenguer M, López-Labrador FX, Wright TL. (2001) Hepatitis C and liver transplantation. J Hepatol 35: 666–678.
5. Powers KA, Ribeiro RM, Patel K, Pianko S, Nyberg L, et al. (2006) Kinetics of hepatitis C virus reinfection after liver transplantation. Liver Transpl 12: 207–216.
6. Garcia-Retortillo M, Forns X, Feliu A, Moitinho E, Costa J, et al. (2002) Hepatitis C virus kinetics during and immediately after liver transplantation. Hepatology 35: 680–687.
7. Berenguer M, Ferrell L, Watson J, Prieto M, Kim M, et al. (2000) HCV-related fibrosis progression following liver transplantation: increase in recent years. J Hepatol 32: 673–684.
8. Yilmaz N, Shiffman ML, Stravitz RT, Sterling RK, Luketic VA, et al. (2007) A prospective evaluation of fibrosis progression in patients with recurrent hepatitis C virus following liver transplantation. Liver Transpl 13: 975–983.
9. Forman LM, Lewis JD, Berlin JA, Feldman HI, Lucey MR. (2002) The association between hepatitis C infection and survival after orthotopic liver transplantation. Gastroenterology 122: 889–896.
10. Thuluvath PJ, Krok KL, Segev DL, Yoo HY. (2007) Trends in post-liver transplant survival in patients with hepatitis C between 1991 and 2001 in the United States. Liver Transpl 13: 719–724.
11. Terrault NA, Berenguer M. (2006) Treating hepatitis C infection in liver transplant recipients. Liver Transpl 12: 1192–1204.
12. Berenguer M, Palau A, Fernandez A, Benlloch S, Aguilera V, et al. (2006) Efficacy, predictors of response, and potential risks associated with antiviral therapy in liver transplant recipients with recurrent hepatitis C. Liver Transpl 12: 1067–1076.
13. Bizollon T, Pradat P, Mabrut JY, Radenne S, Ducerf C, et al. (2007) Histological benefit of retreatment by pegylated interferon alfa-2b and ribavirin in patients with recurrent hepatitis C virus infection post transplantation. Am J Transplant 7: 448–453.
14. Wang CS, Ko HH, Yoshida EM, Marra CA, Richardson K. (2006) Interferon-based combination anti-viral therapy for hepatitis C virus after liver transplantation: a review and quantitative analysis. Am J Transplant 6: 1586–1599.
15. Carrion JA, Navasa M, Garcia-Retortillo M, Garcia-Pagan JC, Crespo G, et al. (2007) Efficacy of antiviral therapy on hepatitis C recurrence after liver transplantation: a randomized controlled study. Gastroenterology 132: 1746–1756.
16. Tamura S, Sugawara Y. (2008) Treatment strategy for hepatitis C after liver transplantation. J Hepatobiliary Pancreat Surg 15: 111–123.
17. Sugawara Y, Makuuchi M, Matsui Y, Kishi Y, Akamatsu N, et al. (2004) Preemptive therapy for hepatitis C virus after living-donor liver transplantation. Transplantation 78: 1308–1311.
18. Tamura S, Sugawara Y, Yamashiki N, Kaneko J, Kokudo N, et al. (2010) Pre-emptive antiviral therapy in living donor liver transplantation for hepatitis C: observation based on a single-center experience. Transpl Int 23: 580–588.
19. Ge D, Fellay J, Thompson AJ, Simon JS, Shianna KV, et al. (2009) Genetic variation in IL28B predicts hepatitis C treatment-induced viral clearance. Nature 461: 399–401.

20. Tanaka Y, Nishida N, Sugiyama M, Kurosaki M, Matsuura K, et al. (2009) Genome-wide association of IL28B with response to pegylated interferon-alpha and ribavirin therapy for chronic hepatitis C. Nat Genet 41: 1105–1109.
21. Suppiah V, Moldovan M, Ahlenstiel G, Berg T, Weltman M, et al. (2009) IL28B is associated with response to chronic hepatitis C interferon-alpha and ribavirin therapy. Nat Genet 41: 1100–1104.
22. Fukuhara T, Taketomi A, Motomura T, Okano S, Ninomiya A, et al. (2010) Variants in IL28B in liver recipients and donors correlate with response to peg-interferon and ribavirin therapy for recurrent hepatitis C. Gastroenterology 139: 1577–1585.
23. Kawaoka T, Takahashi S, Takaki S, Hiramatsu A, Waki K, et al. (2012) Interleukin-28B single nucleotide polymorphism of donors and recipients can predict viral response to pegylated interferon/ribavirin therapy in patients with recurrent hepatitis C after living donor liver transplantation. J Gastroenterol Hepatol 27: 1467–1472.
24. Enomoto N, Sakuma I, Asahina Y, Kurosaki M, Murakami T, et al. (1996) Mutations in the nonstructural protein 5A gene and response to interferon in patients with chronic hepatitis C virus 1b infection. N Engl J Med 334: 77–81.
25. Akuta N, Suzuki F, Sezaki H, Suzuki Y, Hosaka T, et al. (2005) Association of amino acid substitution pattern in core protein of hepatitis C virus genotype 1b high viral load and non-virological response to interferon-ribavirin combination therapy. Intervirology 48: 372–380.
26. Honda M, Sakai A, Yamashita T, Nakamoto Y, Mizukoshi E, et al. (2010) Hepatic ISG expression is associated with genetic variation in interleukin 28B and the outcome of IFN therapy for chronic hepatitis C. Gastroenterology 139: 499–509.
27. Dill MT, Duong FH, Vogt JE, Bibert S, Bochud PY, et al. (2011) Interferon-induced gene expression is a stronger predictor of treatment response than IL28B genotype in patients with hepatitis C. Gastroenterology 140: 1021–1031.
28. Bochud PY, Bibert S, Negro F, Haagmans B, Soulier A, et al. (2011) IL28B polymorphisms predict reduction of HCV RNA from the first day of therapy in chronic hepatitis C. J Hepatol 55: 980–988.
29. Charlton MR, Thompson A, Veldt BJ, Watt K, Tillmann H, et al. (2011) Interleukin-28B polymorphisms are associated with histological recurrence and treatment response following liver transplantation in patients with hepatitis C virus infection. Hepatology 53: 317–324.
30. Duarte-Rojo A, Veldt BJ, Goldstein DD, Tillman HL, Watt KD, et al. (2012) The course of posttransplant hepatitis C infection: comparative impact of donor and recipient source of the favorable IL28B genotype and other variables. Transplantation 94: 197–203.
31. Lange CM, Moradpour D, Dochring A, Lehr HA, Müllhaupt B, et al. (2011) Impact of donor and recipient IL28B rs12979860 genotypes on hepatitis C virus liver graft reinfection. J Hepatol 55: 322–327.
32. Eurich D, Boas-Knoop S, Ruehl M, Schulz M, Carrillo ED, et al. (2011) Relationship between the interleukin-28b gene polymorphism and the histological severity of hepatitis C virus-induced graft inflammation and the response to antiviral therapy after liver transplantation. Liver Transpl 17: 289–298.
33. Crespo G, Carrión JA, Coto-Llerena M, Mariño Z, Lens S, et al. (2013) Combinations of simple baseline variables accurately predict sustained virological response in patients with recurrent hepatitis C after liver transplantation. J Gastroenterol 48: 762–769.
34. Coto-Llerena M, Pérez-Del-Pulgar S, Crespo G, Carrión JA, Martínez SM, et al. (2011) Donor and recipient IL28B polymorphisms in HCV-infected patients undergoing antiviral therapy before and after liver transplantation. Am J Transplant 11: 1051–1057.
35. Thompson AJ, Muir AJ, Sulkowski MS, Ge D, Fellay J, et al. (2010) Interleukin-28B polymorphism improves viral kinetics and is the strongest pretreatment predictor of sustained virologic response in genotype 1 hepatitis C virus. Gastroenterology 139: 120–129.
36. Toyoda H, Kumada T, Tada T, Hayashi K, Honda T, et al. (2012) Predictive value of early viral dynamics during peginterferon and ribavirin combination therapy based on genetic polymorphisms near the IL28B gene in patients infected with HCV genotype 1b. J Med Virol 84: 61–70.
37. Poordad F, McCone J Jr, Bacon BR, Bruno S, Manns MP, et al. (2011) Boceprevir for untreated chronic HCV genotype 1 infection. N Engl J Med 364: 1195–1206.
38. Sherman KE, Flamm SL, Afdhal NH, Nelson DR, Sulkowski MS, et al. (2012) Response-guided telaprevir combination treatment for hepatitis C virus infection. N Engl J Med 365: 1014–1024.
39. Jacobson IM, McHutchison JG, Dusheiko G, Di Bisceglie AM, Reddy KR, et al. (2011) Telaprevir for previously untreated chronic hepatitis C virus infection. N Engl J Med 364: 2405–2416.
40. McCaughan GW. (2012) New therapies against HCV: expected risks and challenges associated with their use in the liver transplant setting. J Hepatol 57: 1361–1367.

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Acute Hepatitis C in HIV-1 Infected Japanese Cohort: Single Center Retrospective Cohort Study

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Abstract

Objectives: HCV co-infection is a poor prognostic factor in HIV-1-infected patients. Although the number of newly reported patients who show seroconversion is increasing, the clinical features are still unclear, especially in Asian countries.

Design: A single-center retrospective cohort study of patients diagnosed between 2001–2012.

Methods: Acute hepatitis C (AHC) was diagnosed upon detection of high serum ALT (>100 IU) followed by anti-HCV seroconversion. Clinical characteristics, HIV-1-related immunological status and IL-28B genotypes (rs12979860, rs8099917) were collected. We compared these variables between patients with and without spontaneous clearance of HCV and between responders and non-responders to treatment with pegylated interferon (PEG-IFN) plus ribavirin.

Results: Thirty-five patients were diagnosed with AHC during the study period. The majority (96.9%) were MSM. Three were lost to follow-up. Seventy-five percent of patients with AHC (24/32) were asymptomatic and found incidentally to have high serum ALT. Compared to those who did not show spontaneous clearance, patients with spontaneous HCV viral clearance showed more symptoms and more severe abnormalities related to acute hepatitis. Spontaneous clearance was seen in 4 out of 28 patients with CC+TT genotype, but not in 6 patients with IL-28B CT+TG genotype. PEG-IFN plus ribavirin treatment was initiated in 12 out of 28 cases without spontaneous clearance. The sustained virological response rate was high (81.8%, 9/11), even in cases with CT+TG genotype infected with HCV genotype 1b (SVR 2/2).

Conclusions: Careful attention to AHC is needed in HIV-1-infected MSM. Early diagnosis and PEG-IFN plus ribavirin treatment should be considered for AHC cases.

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Data Availability: The authors confirm that all data underlying the findings are fully available without restriction. All data supporting our conclusions are included within the manuscript. Original data of our retrospective analyses are available in medical records of our hospital.

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Introduction

The estimated worldwide prevalence of hepatitis C virus (HCV) infection is 2–3% [1]. HCV co-infection increases morbidity rate in HIV infected individuals, and previous meta-analysis reported mortality among patients co-infected with HCV was 1.35 times higher than that among patients with HIV-infection alone even in the highly active antiretroviral therapy (HAART) era [2]. In HIV-1/HCV co-infected patients, progression to liver cirrhosis and hepatocellular carcinoma (HCC) is faster than that in patients without HIV-1 infection [3]. Furthermore, the response to treatment with pegylated interferon (PEG-IFN) plus ribavirin (RBV) in HIV-positive patients with chronic HCV infection is poor (sustained virological response: SVR 19–40%), compared with patients infected with HCV alone (SVR 54–61%) [4–9].

The risk of HCV acquisition via heterosexual intercourse is estimated to be very low [10]. Recently, however, a high incidence of HCV seroconversion has been reported in HIV-1 infected men who have sex with men (MSM) [11–13]. These results suggest that new HCV infection can be a potential future problem in the clinical management of HIV-1 infected patients. On the other hand, a favorable response to treatment with PEG-IFN plus RBV for acute hepatitis C (AHC) relative to that for chronic one has been reported in HCV-infected (SVR 85–98%) [14,15] and HIV/HCV co-infected patients (SVR 60–80%) [16,17]. In this regard, the recent guidelines recommend PEG-IFN plus RBV treatment