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<th>HB Vaccination in the Prevention of Viral Reactivation in Allogeneic Hematopoietic Stem Cell Transplantation Recipients with Previous HBV Infection</th>
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<td>Author(s)</td>
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**HB vaccine to prevent viral reactivation in allogeneic hematopoietic stem cell transplantation recipients with previous HBV infection.**

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List of Abbreviations

HBV, hepatitis B virus
RS, reverse seroconversion
HSCT, hematopoietic stem cell transplantation
HSV, herpes simplex virus
VZV, varicella zoster virus
HHV6, human herpes virus 6
EBV, Epstein-barr virus
CMV, cytomegalovirus
PCR, polymerse chain reaction
Anti-HBs, anti-hepatitis B surfase antigen antibody
HBsAg, hepatitis B antigen
Anti-HBc, anti-hepatitis B core antigen antibody
HBIG, hepatitis B immunoglobulin

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Abstract

HBV-reverse seroconversion (RS) following allogeneic hematopoietic stem cell transplantation (allo-HSCT) is a frequent late-onset complication in recipients with previous HBV infection. We conducted post-transplant HB vaccine intervention in 38 allo-HSCT recipients with previous HBV infection. Firstly, we followed the recipients without any intervention (historical control) until 2003; hence we commenced HB vaccination. Out of the patients who underwent transplantation after 2003, 13 recipients were immunized by a standard 3-dose regimen after immunosuppressant cessation (vaccine group), while 12 recipients were observed without any intervention (non-vaccine group). Eight of the 13 historical control group recipients and 3 of the 12 non-vaccine group recipients, but none of the 13 vaccine group recipients, suffered HBV-RS. Cumulative risks of HBV-RS at 3 years post-HSCT in the historical control, non-vaccine and vaccine groups were 41%, 39% and 0% respectively (P=0.022). We therefore conclude that intervention with HB vaccines is significantly effective in preventing post-HSCT HBV-RS.
Introduction

Hematopoietic stem cell transplantation (HSCT) is performed worldwide to treat various diseases, including hematological malignancies. Intensive chemotherapy and immunosuppression, in patients who undergo HSCT, result in immune dysfunction putting these patients at high risk for infection, not only with external pathogens but also internal ones. Reactivation of herpes viruses such as Herpes simplex (HSV), Varicella zoster (VZV), Human Herpes Virus 6 (HHV6), Epstein-Barr (EBV), and Cytomegalovirus (CMV), for instance, are well-known phenomena in such patients; recently hepatitis B virus (HBV) has also been recognized to be reactivated following HSCT.1-4

The appearance of anti-hepatitis B surface antigen antibody (anti-HBs) and the clearance of HBV from the serum usually indicate resolution of hepatitis in patients infected with HBV. However, most patients in whom HBV has been eliminated from the serum still have HBV DNA in the liver that is detectable by polymerase chain reaction (PCR).5 Reactivation of dormant HBV in the liver has been observed in an immunocompromised status, such as HSCT, renal transplantation, intensive chemotherapy or rituximab use.6-13 The reactivation of hepatitis in anti-HBs positive patients is known as reverse seroconversion (RS). Previously, we have revealed that RS is a late-onset complication with high frequency that can be predicted by careful monitoring of the progressive disappearance of anti-HBs.1 From earlier observations, we consider that RS hepatitis after HSCT is a hepatitis caused by reconstituted naïve donor immunity, following the loss of recipient-derived immunity against HBV.

In the current study, we conducted post-transplant recombinant HB (rHB) vaccine intervention to immunize naïve donor immunity and to determine its clinical efficacy in preventing post-transplant HBV-RS.
Patients and Methods

Patients Studied. Allo-HSCT recipients who underwent transplantation in our hospital, from February 1990 to March 2007, and who were followed for at least 1 year after HSCT, were enrolled in this study. We retrospectively studied 38 recipients with previous HBV infection in the pre-HSCT evaluation. We started hepatitis B vaccine intervention in HSCT recipients after cessation of immunosuppressant administration, in March 2003. Thirteen recipients, who underwent transplantation after March 2003, were immunized with HB vaccine using a standard regimen (vaccine group). Twelve recipients, who also underwent transplantation after March 2003, were observed without intervention, either due to prolonged administration of immunosuppressant or due to discontinuation of follow-up within our hospital (non-vaccine group). Data regarding the progress of these patients were obtained through collaboration with other hospitals in the region. Thirteen recipients who underwent transplantation before March 2003, and were observed without vaccine intervention, were considered as controls (historical control group). We studied the transition of anti-HBs and the incidence of HBV-RS. Patients’ characteristics are shown in Table 1. Reflecting the historical background, the number of RIST and median age were higher in vaccine and non-vaccine groups compared to the historical control group. All variables are equivocal between the vaccine and non-vaccine groups. This study was approved by the institutional review board of the Hokkaido University School of Medicine.

Definitions. Previous HBV infection was diagnosed serologically. Recipients with [HBsAg (-), anti-HBs (+), anti-HBc (+)] and [HBsAg(-), anti-HBs (-), anti-HBc (+)] were both considered to have had previous HBV infection. In patients with [HBsAg (-), anti-HBs (+), anti-HBc (-)] we diagnosed previous HBV infection only after confirming that they had never received rHB vaccination, since normally rHB vaccination responders show the same
serological patterns. RS was defined as the disappearance of anti-HBs and the appearance of HBsAg and HBV-DNA in the serum, irrespective of the presence of clinical hepatitis. From 2003, HSCT recipients who were serologically diagnosed to have had previous HBV infection were immunized with rHB vaccine following cessation of immunosuppressants. One cycle consisted of a 3 dose-schedule, given at 0, 1 and 6 months. Yeast-derived rHB vaccines containing the major surface protein (Bimmugen, Astellas Pharmaceutical, Tokyo; Heptavax-II, Merck & Co., Whitehouse Station) were used. Vaccine responders were defined with anti-HBs levels >10 mIU/ml, which are considered to be protective. In patients whose anti-HBs titer was already >10 mIU/ml at the pre-vaccination point, vaccine responders were determined by their tendency to increase anti-HBs levels after vaccination.

Results and Discussion

Progressive decreases in anti-HBs titers were observed in all pre-HSCT anti-HBs-positive recipients. Four of the 13 recipients (31%) responded to initial courses of the HB vaccine. Three of the 4 vaccine responders responded after the third vaccine (Figure 1A). Eight of the 13 historical control group recipients and 3 of the 12 non-vaccine group recipients suffered HBV-RS following loss of anti-HBs, but none of the 13 vaccine group recipients suffered HBV-RS (7 of the 8 patients having HBV-RS in the historical control group were reported in our previous paper¹). Only 1 of the 8 historical control group recipients, and none of the 3 of non-vaccine group recipients, suffered HBV-RS during immunosuppressant administration, thus meaning that the use of immunosuppressant's did not significantly affect the occurrence of HBV-RS. Cumulative risks of HBV-RS at 3 years post-HSCT in the historical control, non-vaccine group and vaccine group were 41%, 39%
and 0% respectively (P=0.022) (Figure 1B).

Although patients with HBsAg have clearly been shown to be a high-risk group for liver complications, little attention has been paid to patients having anti-HBs when performing HSCT. In Japan, the population of patients having had previous BV infection (15-20%) is more than 10 times higher than that of HBsAg-positive carriers (1%). In patients with resolved HBV infection, the progressive disappearance of anti-HBs is inevitable; occurring with the progressive loss of recipient-type immune cells, regardless of the pre-transplantation anti-HBs titer. Thus, patients with resolved HBV infection should be considered to be a high risk group for HBV reactivation because of the persistence of the virus in a “latent” state in the liver. HBV reactivation from dormant HBV, which remains in the liver after the initial infection, was first recognized in the liver transplantation field. Uemoto S et al. reported that out of 16 recipients with no prior history of HBV infection, who underwent liver transplantation from anti-HBc-positive and HbsAg-negative living donors, 15 (93.75%) became HBsAg-positive after transplantation. Apart from proving the existence of HBV-DNA in liver tissues, even after serological resolution of HBV infection, they also discovered that HBV is transmitted to recipients by liver grafts from anti-HBc-positive donors at a significantly high rate. In the liver transplantation field, the use of a preventive measure, which basically entails creating passive immunity by means of hepatitis B immunoglobulin (HBIG) and lamivudine, has already been established. However, due to the limited commercial supply of HBIG as well as the lifelong requirement for prophylaxis following transplantation, financial constraints represent a debilitating problem to this therapy. Moreover, despite the use of rHB vaccination to reduce the frequency of HBIG support, active immunization showed limited effect under sustained usage of immunosuppressants.
RS of HBV following HSCT, and HBV reactivation following liver transplantation from donors with previous HBV infection are reciprocal phenomena. The most important difference between solid organ transplants and HSCT is the span of immunosuppressant usage. In solid organ transplants immunosuppressant administration is lifelong but, in HSCT, immunity is reconstituted and treatment with immunosuppressants can be stopped. We therefore hypothesized that active immunization by rHB vaccine might be useful in avoiding HBV-RS after HSCT. The safety and efficacy of rHB vaccination in HSCT recipients has already been reported. Some guidelines for post-transplant vaccination recommend rHB vaccination, mainly to prevent de novo infection of HBV from outside the body. It was the European Group for Blood and Marrow Transplantation (EBMT), which for the first time, proposed the use of rHB vaccination to prevent innate HBV reactivation as a post-transplant vaccination option. The clinical efficacy of the rHB vaccine in preventing post-transplant HBV-RS, however has not been previously reported. Notwithstanding that this study is a non-randomized historical case-control study, this is the first report in which the usefulness of HB vaccine in preventing HBV-RS after HSCT has been investigated.

Several studies have recommended prophylactic vaccination of the donor, expecting adoptive transfer of HBV immunity; a well-described phenomenon in the literature, regrettably however, attempts to overcome immunodeficiency by immunizing the donor have not always been successful. Long-term immunity, defined as the persistence of antibody presence, is not achieved without re-exposure to the specific antigen, either by re-immunization or re-infection, regardless of the immune status of the donor. In our study, the rate of successful vaccinations was lower than that described in a previous report. Interestingly, however, in our study 4 of the 9 non-responders (44%) showed a slight increase in their anti-HBs titer after HB vaccination, even though the responder
criteria (>10 mIU/ml) were not met. Consequently, in non-responders, an intensified dose or schedule, possibly using another rHB vaccine should be taken into consideration.

In conclusion, RS is a late-onset complication with high frequency following HSCT, which can be predicted by careful monitoring of the progressive disappearance of anti-HBs. In view of the results obtained in this study we propose rHB vaccination of recipients as a valuable prophylactic tool for the reactivation of HBV in allo-HSCT patients.

Acknowledgements

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This work was supported by Grants-in-Aid for Scientific Research (KAKENHI) (No. 20790667) from the Japan Society for the Promotion of Science (JSPS). S.D. is a Research Fellow of the Japan Society for the Promotion of Science.
References


Figure legends

Table 1. Patients’ characteristics

MAC = myeloablative conditioning; RIST = reduced intensity conditioning;
MTX = methotrexate; CSA = cyclosporin A; FK506 = tacrolimus
GVHD = graft versus host disease

Figure 1.
(A) Immunization effect of HB vaccination.
Transition of anti-HBs in the ‘vaccine group’ from the pre-HSCT to the pre-vaccination point, and from the pre-vaccination point onwards, is plotted. The first vaccine started at 6-29 months (median 17 months) after HSCT. In 8 of the 13 pre-HSCT-anti-HBs-positive recipients, the anti-HBs titer had already decreased to undetectable levels at the time of the 1st vaccine. Although 8 out of the 13 recipients (fine line) showed an increase in anti-HBs, only 4 recipients (bold line) showed an increase of anti-HBs levels >10 mIU/ml. Non-responders are depicted with dotted lines in the graph.

(B) Cumulative incidence of post-transplant HBV-RS.
The primary endpoint was the occurrence of RS. The actual risks of the endpoints were estimated using the Kaplan-Meier method. Survival curves were compared using the log-rank test. There were no differences between the ‘Historical control group’ and the ‘Non-vaccine group’ (P=0.74). The ‘Vaccine group’ showed a significantly lower risk for HBV-RS than the other two groups (P=0.022).
Table 1. Patients’ characteristics

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<tr>
<td>Male / Female</td>
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Figure 1A. Immunization effect of HB vaccination

Anti-HBs (mIU/ml)

Pre-HSCT  Pre-vaccination  Post-1st vaccination  Post-2nd vaccination  Post-3rd vaccination
Figure 1B. Cumulative incidence of post-transplant HBV-RS

(%) 100 90 80 70 60 50 40 30 20 10 0

Historical control (n=13)

Non-vaccine group (n=12)

Vaccine group (n=13)

P=0.74

P=0.022

Months after allo-HSCT