RESEARCH NOTE

SEROLOGICAL RESPONSE TO DOUBLE-DOSE HEPATITIS B RE-VACCINATION IN HIV-INFECTED ADOLESCENTS

Pagakrong Lumbiganon[1], Pope Kosalaraksa[1] and Thanitta Udomphanit[2]

[1]Department of Pediatrics, [2]Srinagarind Hospital, Faculty of Medicine, Khon Kaen University, Khon Kaen, Thailand

Abstract. The aim of this study was to determine serological response to double-dose hepatitis B re-vaccination among HIV-infected adolescents who did not response to re-vaccination with standard-dose hepatitis B vaccine. We enrolled adolescents ≥12 to 18 years of age who had serological negative hepatitis B surface antigen, hepatitis B surface (anti-HBs) and hepatitis B core antibodies at screening and after earlier re-vaccination with three standard-doses of hepatitis B vaccine still had negative anti-HBs (<10 IU/l). Double-dose hepatitis B vaccine (<16 years of age: 20 μg; ≥16 years of age: 40 μg) was given at month 0 and 3. Anti-HBs was tested at month 3 along with the second double-dose. Patients with negative anti-HBs were given a 3rd dose at month 6 and presence of antibodies was tested at month 9. Of the 25 patients enrolled (10 males, 15 females; 17 = 12-15 and 8 =16-18 years of age), the median (interquartile range) CD4 level at enrollment was 635 (427-766) cells/mm^3, with 22 on highly active antiretroviral therapy (HAART) and 20/23 tested had suppression of HIV load. Anti-HBs were positive in 21 patients after the first dose, of whom 20 were on HAART and 18 had viral suppression. Among four non-responders (after the first dose), two were not on HAART and two were on HAART with HIV viral suppression. This study demonstrates that double-dose hepatitis B re-vaccination can result in serological responses in a majority of previously non-responsive HIV-infected adolescents.

Keywords: hepatitis B vaccine, double-dose re-vaccination, adolescents, HIV

Correspondence: Pagakrong Lumbiganon, Department of Pediatrics, Faculty of Medicine, Khon Kaen University, Khon Kaen 40002, Thailand.
Tel: +66 (0) 43 348382; Fax: +66 (0) 43 348382
E-mail: paglum@kku.ac.th

This study was presented in part as a poster at the 9th World Congress on Pediatric Infectious Diseases, Rio De Janeiro, Brazil, 18-21 November 2015.

INTRODUCTION

HIV-infected patients have a higher risk of chronic infection after hepatitis B virus exposure, and have a faster progression to cirrhosis and hepatitis B-related complication than HIV-negative individuals (Panel on Opportunistic Infections in HIV-Exposed and HIV-Infected Children, 2013). Hepatitis B immunization is es-

sential in all patients with HIV-infection to prevent liver-related morbidity and mortality, which can occur with hepatitis B virus co-infection. However, both HIV-infected children and adults have low immunological responses (17-24%) to standard dose hepatitis B vaccination compared to responses of >90% in HIV-uninfected individuals (Siriaksorn *et al*, 2006; Landrum *et al*, 2009). A previous study at four pediatric HIV clinics in Thailand showed in 521 HIV-infected adolescents, ≥12-25 years of age, protective hepatitis B surface antibodies (anti-HBs) level ≥10 IU/l is present in only 18% of the subjects although 97% were born after 1992 when routine hepatitis B immunization since birth was implemented in this country (Aurpibul *et al*, 2012).

Re-vaccination after highly active antiretroviral therapy (HAART) and immune reconstitution has been recommended for patients lacking immunity to hepatitis B (Panel on Opportunistic Infections in HIV-Exposed and HIV-Infected Children, 2013). A study from Thailand found antibody response to three standard-dose hepatitis B re-vaccination in 63 HIV-infected children with immune recovery on HAART of 92% (Lao-Araya *et al*, 2007). However, for those who do not respond to the standard-dose hepatitis B re-vaccination, there is no consensus guideline for further re-vaccination. Improved serological response rates to larger doses or increased numbers of doses of hepatitis B vaccine in adults (Potsch *et al*, 2012; Crum-Cianflone and Wallace 2015) and in HIV-infected children and adolescents (Flynn *et al*, 2011; Bunupuradah *et al*, 2013) have been reported. On the other hand, a multicenter open-label, randomized trial in France involving 176 HIV-infected adults found no difference in proportion of anti-HBs responders among those who received double-dose compared with those who received standard-dose vaccine although the double-dose group achieved higher anti-HBs titer (Rey *et al*, 2015).

HIV-infected adolescents are at risk of acquiring hepatitis B virus infection through high-risk behavior, such as unsafe sex practice and use of injected drugs of abuse. Co-infection with hepatitis B virus is responsible for high morbidity and mortality among HIV-infected adults (Panel on Opportunistic Infections in HIV-Exposed and HIV-Infected Children, 2013). Hence, this study determined serological response to double-dose hepatitis B re-vaccination in HIV-infected adolescents who did not response to standard-dose hepatitis B re-vaccination.

MATERIALS AND METHODS

Subjects

The study was conducted at the pediatric HIV Clinic, Srinagarind Hospital, Faculty of Medicine, Khon Kaen University, Khon Kaen, Thailand. HIV-infected adolescents ≥12 - 18 years of age who had negative hepatitis B surface antigen (HBsAg), negative anti-HBs and hepatitis B core antibody (anti-HBc) measured by chemiluminescent microparticle immunoassay (CMIA), but had a history of hepatitis B vaccination since birth or were born after 1992 when the universal hepatitis B immunization was implemented in Thailand were given three standard-doses of hepatitis B vaccine. Anti-HBs was tested three months after the 3rd dose. Patients with anti-HBs titers <10 IU/l were invited to participate in this double-dose study.

The study was approved by Khon Kaen University Ethics Committee for Human Research (HE 561435). Before enrollment, written informed consent was

obtained from participant or from parent or legal custodian for participants <18 years of age.

Vaccination protocol and laboratory assays

Hepatitis B vaccine (Government Pharmaceutical Organization-Merieux Biological Products, Bangkok, Thailand) containing 20 μg of purified HBsAg and 0.5 mg of aluminum hydroxide (as adjuvant) (recommended dose for person at least 16 years old) was used. Double-dose vaccine (<16 years old: 20 μg; ≥16 years old: 40 μg) was given at month 0 and 3 intramuscularly at the deltoid area. For the 40 μg dose, each vaccine injection was given separately into each arm. Anti-HBs antibody was evaluated by CMIA at month 3 after the first dose along with the second double-dose of hepatitis B vaccine. Only patients who had anti-HBs <10 IU/l were given a 3rd dose of hepatitis B vaccine at month 6 and anti-HBs was re-tested at month 3 after the 3rd dose of vaccine. The 3-month interval was chosen to coincide with the patients' regular HIV clinic visits.

CD4 level was determined by flow cytometry (3- color immunophenotyping) and viral load by real time PCR.

Patients' baseline characteristics are reported as median with interquartile ranges (IQR) and ranges or frequencies as appropriate. Values closest (within +/-three months) to the time of first double-dose hepatitis B re-vaccination were used. Percent serological response rate were calculated.

Vaccine safety and tolerability were assessed by asking patients and caregivers at each three-monthly post re-vaccination visit regarding side effects, such as high fever, rash, and severe pain at the injection site occurring during the two weeks after each vaccination. Patients or caregivers were asked to immediately notify the study nurse by phone whenever any severe reaction occurred.

RESULTS

From February 2014 to June 2015, 25 patients (10 males and 15 females) were enrolled in the study. Seventeen (68%) participants were 12-15 and 8 (32%) were 16-18 years of age, with median age of 15 years old (IQR 14 -16) and median body mass index was 19.3 kg/m^2 (IQR 17.9-20.7, range 12.8-25.8). Median CD4 level was 635 cells/mm^3 (IQR 427-766, range 228-1,103) and the median percent CD4 cells was 26.4 (IQR 20.0-30.1, range 14.6-37.5). At baseline, 22 (88%) patients were receiving HAART with median duration of 8.0 years (IQR 6.5-11.5, range 0.6-14.6) and 20 (80%) patients achieved HIV viral suppression (<20 copies/ml) (Table1).

Anti-HBs titers tested month 3 after the first dose were ≥10 IU/ml in 21 (84%) patients, while four (16%) patients had titers <10 IU/ml. The median duration (IQR) of HAART in these two groups was 8.1 (6.9-11.8) and 0.9 (0.6-1.2) years, respectively. Among 21 patients who responded to the first double-dose of vaccine, 15 (71%) had anti-HBs titers >100 IU/l, among whom was a 15-year-old girl who refused to continue HAART due to poor compliance and was on lamivudine alone for six months before the first double-dose vaccination. Her CD4 level was 228 cells/mm^3 (15%) with viral load of 26,367 and she developed an anti-HBs titer of 214.2 IU/l. Among seven patients with anti-HBs antibody titers >1,000 IU/l, six had CD4 level >350 cells/mm^3 and HIV viral suppression. A 14-year old male with a CD4 level of 346 cells/mm^3 (17.5%) and HIV viral suppression (<20 copies/ml) also responded with anti-HBs titer >1,000 IU/l.

Table 1
Baseline patients' characteristics and hepatitis B surface antibody (anti-HBs) response to double-dose hepatitis B vaccination regimen.

Characteristic[a]		Anti-HBs response after 1st double dose vaccination				
		Antibody positive (≥10 IU/l) (n = 21, 84%)				Antibody negative (<10 IU/l) (n = 4)
		Total	<100 IU/l (n = 6)	≥100-999 IU/l (n = 8)	≥1,000 IU/l (n = 7)	
Male:female	2:3	3:4	1:2	3:5	4:3	1:3
Median age (IQR), years	15 (14-16)	15 (15-16)	15 (14-16)	15 (15-17)	15 (15-17)	14 (13-15)
Median (IQR) CD4 cells	26 (20-30)	26 (19-31)	22 (17-29)	28 (22-34)	30 (21-32)	24 (21-28)
Median (IQR) CD4 cell count	635 (427-766)	596 (410-794)	646 (427-794)	537 (375-817)	635 (407-972)	761 (602-764)
CD4 cell count >350 cells/mm^3	23/25	19/21	6/6	7/8	6/7	4/4
Viral load <20 copies/ml	20/23	18/21	4/6	7/8	7/7	2/2[b]
Received HAART	22/25	20/21	6/6	7/8	7/7	2/4
Median (IQR) duration of HAART, years	8.0 (6.5-11.5)	8.1 (6.9-11.8)	7.2 (5.2-8.0)	10.5 (7.8-13.1)	8.2 (7.3-12.0)	0.9 (0.6-1.2)

[a]Value nearest to time of first dose of hepatitis B vaccination (±3 months); [b]One patient never received HAART and another ceased HAART 3 years prior were not tested. HAART, highly active antiretroviral therapy; IQR, interquartile range.

Among four patients who did not achieve anti-HBs titers ≥10 IU/ml after the first double-dose vaccine, all had baseline CD4 cell counts >350 cells/mm^3 and three were female.

Two patients had received HAART for 0.6 and 1.2 years, respectively and had HIV viral suppression with CD4 level of 762 (20%) and 444 cells/mm^3 (29.8%), respectively. The other two patients not on HAART were a 16-year old girl who had been followed-up for 9 years and never received HAART due to persistently high CD4 level (841 cells/mm^3, 27.5%) and a 14-year old boy who had ceased HAART for three years prior to enrollment in this program at his request due to poor compliance and with a CD4 of 764 cells/mm^3 (21.1%). All four patients received the third double-dose of hepatitis B vaccine, and their anti-HBs remained negative month 3 after the third dose.

Three (12%) patients reported minor pain at the injection site after the first dose. None reported any side effects, such as rash, swelling or high fever. After the second and third dose, no adverse event was reported.

DISCUSSION

In this study, the anti-HBs response to a single double-dose hepatitis B vaccine in non-immune HIV-infected adolescents was 84%, with high titers (>100 IU/l) in 71% of responders. Among the non-responder group (n = 4), all had CD4 levels >350 cells/mm^3, and two patients on HAART had viral suppression. There were no adverse events from the double-dose hepatitis B vaccine.

A previous study in Thailand of a double-dose hepatitis B vaccination reported among seven adolescents [mean age (SD) of 15.4 (3.4) years] given at months 0, 1 and 2, six having anti-HBs response of ≥10 IU/l after the first and second dose and all seven one month after the 3rd dose; all patients were on HAART with mean (SD) duration of 6.9 (3.6) years (Bunupuradah *et al*, 2013). In our study, four patients with no response to the full three double-dose series had high CD4 levels, two of whom were not on HAART and the other two had been on HAART for only 0.6 and 1.2 years prior to enrollment. Longer duration of HAART might be a factor associated with good serological response to hepatitis B vaccine.

Our study was limited by the small numbers of eligible adolescents, so a randomized trial was not possible. Measurement of anti-HBs titers after the second and third dose of the double-dose vaccine in patients who developed protective antibody after the first dose was not performed due to limited resources. Nevertheless, evaluation of long term protection in these adolescents is warranted.

In conclusion, double-dose hepatitis B vaccination is safe and could be one of the options for HIV-infected adolescents who have no protective antibody response after a standard-dose hepatitis B vaccination.

ACKNOWLEDGEMENTS

The authors thank the Thailand Universal Coverage Scheme for providing hepatitis B vaccines, participants and families or caregivers for their cooperation and Dr Annette H Sohn for reviewing the manuscript.

CONFLICTS OF INTEREST

The authors declare no conflicts of interest.

REFERENCES

Aurpibul L, Lumbiganon P, Kolasaraksa P, *et al*.

HIV and hepatitis B coinfection among perinatally HIV-infected Thai adolescents. *Pediatr Infect Dis J* 2012; 31: 943-7.

Bunupuradah T, Ananworanich J, Puthanakit T. Double-dose hepatitis B revaccination in nonresponsive HIV-infected adolescents. *J Int Assoc Provid AIDS Care* 2013; 12: 157-8.

Crum-Cianflone NF, Wallace MR. Double-dose hepatitis B vaccination in people with HIV. *Lancet Infect Dis* 2015; 15: 1245-6.

Flynn PM, Cunningham CK, Rudy B, *et al*; Adolescent Medicine Trials Network for HIV/AIDS Interventions (ATN). Hepatitis B vaccination in HIV-infected youth: a randomized trial of three regimens. *J Acquir Immune Defic Syndr* 2011; 56: 325-32.

Landrum ML, Huppler Hullsiek K, Ganesan A, *et al*. Hepatitis B vaccine responses in a large U.S. military cohort of HIV-infected individuals: another benefit of HAART in those with preserved CD4 count. *Vaccine* 2009; 27: 4731-8.

Lao-Araya M, Puthanakit T, Aurpibul L, Sirisanthana T, Sirisanthana V. Antibody response to hepatitis B re-vaccination in HIV-infected children with immune recovery on highly active antiretroviral therapy. *Vaccine* 2007; 25: 5324-9.

Panel on Opportunistic Infections in HIV-Exposed and HIV-Infected Children. Guidelines for the prevention and treatment of opportunistic infections in HIV-exposed and HIV-infected children. Washington, DC: Department of Health and Human Services, 2013. [Cited 2016 Jul 13]. Available from: http://aidsinfo.nih.gov/contentfiles/lvguidelines/oi_guidelines_pediatrics.pdf

Potsch DV, Camacho LA, Tuboi S, *et al.* Vaccination against hepatitis B with 4-double doses increases response rates and antibodies titers in HIV-infected adults. *Vaccine* 2012; 30: 5973-7.

Rey D, Piroth L, Wendling MJ, *et al;* ANRS HB04 B-BOOST Study Group. Safety and immunogenicity of double-dose versus standard-dose hepatitis B revaccination in non-responding adults with HIV-1 (ANRS HB04 B-BOOST): a multicentre, open-label, randomised controlled trial. *Lancet Infect Dis* 2015; 15: 1283-91.

Siriaksorn S, Puthanakit T, Sirisanthana T, Sirisanthana V. Prevalence of protective antibody against hepatitis B virus in HIV-infected children with immune recovery after highly active antiretroviral therapy. *Vaccine* 2006; 24: 3095-9.