ORIGINAL ARTICLE

Hepatitis B seroprevalence among University of Malaya Students in the Post-universal Infant Vaccination Era

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SUMMARY

Aim: A nationwide HBV vaccination for neonates in the Expanded Programme on Immunization (EPI) was implemented in Malaysia in 1989. The objective of this study was to investigate the prevalence of HBsAg, anti-HBs and anti-HBc among the new student intakes in the Faculties of Medicine and Dentistry, University of Malaya from 2005 to 2011.

Materials and Methods: All new students enrolled for undergraduate and postgraduate courses were screened for HBV infection. Serum samples collected were tested for the presence of HBsAg, anti-HBs and anti-HBc with the use of fully automated analysers. Statistical analyses were done using Open Epi version 2.3.1

Results: The overall HBsAg prevalence among the 2923 new intakes was 0.62%. The HBsAg prevalence rate was 1.08% (15/1390) for those born before 1989 and only 0.20% (3/1533) among those born in or after 1989. By year of testing, HBsAg prevalence declined steadily from 1.27% (5/394) in 2005 to 1.20% (5/418) in 2006, 0.95% (4/421) in 2007, 0.49% (2/410) in 2008, 0.49% (2/407) in 2009 and finally 0% in both 2010 (0/445) and 2011 (0/428). Although 66.14% of those vaccinated during infancy had no demonstrable immunity at the time of screening, only 6 (0.39%) students were found to have anti-HBc, including the 3 who were HBsAg positive.

Conclusion: These findings suggested effective prevention of HBV transmission with the universal and voluntary vaccination programs in Malaysia.

KEY WORDS:
Hepatitis B virus, EPI, HBsAg, Anti-HBs, Students

INTRODUCTION

Hepatitis B virus (HBV) infection is a major cause of liver pathology presenting as acute hepatitis and chronic complications such as cirrhosis and hepatocelllular carcinoma. Individuals infected perinatally or in early childhood are at a greater risk of progression from acute to chronic infection than those infected later in life. Asia and the Western Pacific region share about 75% of chronic HBV infections in the world [1]. In public health surveillance, persisting HBV infection is detected by the presence of hepatitis B surface antigen (HBsAg) in the blood. Countries with HBsAg prevalence of at least 8% are classified as high endemicity countries [2].

Since the introduction of HBV vaccination in the early 1980s, the prevalence of HBsAg positivity has declined, particularly in hyperendemic areas where transmission is mostly perinatal or during early childhood [2]. The World Health Organization (WHO) through its Global Advisory Group of the Expanded Program on Immunization (EPI), recommended hepatitis B vaccination to be integrated into national immunization programmes in countries with HBsAg prevalence of 8% or more. In the early stages of this extended immunization programme, the global infant HBV vaccine coverage was low and varied, but by 2007 the coverage rate had reached 65% in most countries [3] and data from hyperendemic regions have shown the effectiveness of the programme for the prevention of HBV transmission. In Taiwan, for instance, within ten years of HBV vaccine introduction, the HBsAg prevalence declined from 9.3% to 1.3% in children younger than 12 years [4] and from 9.8% to 0.7% among those younger than 15 years of age [5]. Similarly, in Thailand, HBsAg prevalence among 1 to 18-year-olds fell from 3.4% to 0.7% ten years after the implementation of EPI with HBV vaccination [6].

Malaysia was an intermediate endemicity country with HBsAg prevalence of 5-7% before nationwide HBV vaccination for neonates was introduced in 1989 [7]. Under this EPI, all newborns are given three doses of HBV vaccine to prevent mother-to-infant transmission. The first dose is given in the hospital soon after birth, the second at the one month post- partum follow up visit, and the third, at the fifth month, either in the hospital or at a rural health centre. The impact of this programme on the prevalence of chronic HBV infection in the Malaysian population has been assessed by Ng *et al.*[8] for school children aged 7-12 years. The HBsAg detection rate in these children declined from 2.5% among those born in 1985 (before the implementation of universal infant vaccination) to 0.4% among those born in 1996.

Many universities screen applicants for medical, dental and nursing courses, as healthcare workers are at risk for infection through exposure to patients' body fluids but conversely, if infected, can potentially transmit their HBV to patients under their care. In 1994, the Britain's Committee of Vice-Chancellors and Principals issued guidance to universities on medical and dental students' fitness to practise in relation to

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hepatitis B[9]. Universities that have adopted this committee's guidance have barred HBV-infected individuals from entry into medical and dental courses and from post-graduate specialist training. In regions with high HBV endemicity, however, there has been no consensus on the suitability of students with good academic achievement but infected with HBV, for medical and dental training.

The objective of this study was to investigate the prevalence of HBsAg and anti-HBs among the new student intakes in the Faculties of Medicine and Dentistry, University of Malaya. The anti-HBc, a serological marker of HBV infection, was analyzed among new intakes vaccinated under the EPI, to study the level of post-vaccination exposure to HBV infection.

MATERIALS AND METHODS

This is a prospective cross-sectional study from 2005 to 2011. All new students enrolled for undergraduate and postgraduate courses in the Faculty of Medicine and Faculty of Dentistry were screened for HBV infection. Demographic data and history of HBV vaccination, including the number and timing of vaccine doses given, were obtained from each student, using a standard questionnaire. All students born in and after 1989 were considered as being vaccinated under the EPI. As HBV screening of students is a routine procedure at the University and only anonymous HBV data was used for analysis, ethics committee review for the study was waived in accordance with the standard operating procedure of the university's Medical Ethics Review Committee.

Detection of HBsAg and anti-HBs

Serum samples collected from 2005 to 2007 were tested for the presence of HBsAg and anti-HBs by a fully automated analyser, AxSym Abbott Plus (Abbott Laboratories, Diagnostics Division, Abbott Park, IL USA). From 2008 to 2011, serum samples were analyzed by the Architect i 2000 (Abbott Laboratories, Diagnostics Division, Abbott Park, IL USA). Assay procedures were conducted according to the manufacturer's instructions. All samples found to be HBsAg reactive in the first screen were re-tested in the same analyser and only repeatedly reactive samples were considered HBsAg positive. Anti-HBs was reported as >/=10 mIU/ml.

Detection of Anti-HBc IgG

Anti-HBc IgG was assayed using the Architect Anti-HBc II (Abbott Laboratories, Diagnostics Division, Abbott Park, IL USA) and conducted according to the manufacturer's instructions. This test was only performed for new intakes born in or after 1989 i.e. those who had received HBV vaccination from EPI.

Statistical analyses

Statistical analyses were done using Open Epi version 2.3.1. Descriptive analyses were used for the baseline demographic and clinical data. Bivariate analyses (i.e Student independent-test and Chi-square) were conducted to test the association between demographic and clinical data. P value of <0.05 was used to test the significance level. Odds ratio and its associated 95% confidence interval were used for categorical data analysis to estimate risk.

RESULTS

A total of 2923 new students were enrolled from 2005 to 2010 for Medicine (47.35%), Dentistry (18.10%), Pharmacy (15.60%), Biomedical Sciences (9.54%), Nursing Degree program (8.01%) and Bio-imaging (1.40%). The male to female ratio was 1:2.4 and the ethnic composition was 55.11% Malay, 39.21% Chinese, 4.41% Indian and 1.27% of other ethnic groups (Table I).

The HBsAg prevalence was 0.62% overall, 1.08% (15/1390) among those born before 1989 and only 0.20% (3/1533) among those born in or after 1989 (Table II). By year of testing, HBsAg prevalence declined steadily from 1.27% (5/394) in 2005 to 1.20% (5/418) in 2006, 0.95% (4/421) in 2007, 0.49% (2/410) in 2008, 0.49% (2/407) in 2009 and finally 0% in both 2010 (0/445) and 2011 (0/428).

Among the 18 students found to be HBsAg positive, there were 15 females, 10 Chinese and 8 Malays, giving an overall seroprevalence of 0.73% in females, 0.35% in males, 0.87% in Chinese and 0.50% in Malays. These results are consistent with previous observations of higher HBV prevalence among females and Chinese in the general Malaysian population (University Malaya data-on-file). By course of study, the overall HBsAg prevalence from 2005 to 2011 was 0.29% (4/1384), 0.57% (3/529), 0.88% (4/456), 1.08% (3/279) and 1.71% (4/234) for Medicine, Dentistry, Pharmacy, Biomedical Sciences and Nursing respectively. The high prevalence among nurses is probably due to their long years of hospital service before they enrolled into the Nursing Degree Programme.

As the Malaysian National EPI was implemented in 1989, students were categorised into a) the EPI group if they were born in or after 1989 and were vaccinated at birth, b) the non-EPI group if they were born before 1989 and had a history of voluntary vaccination and c) the not vaccinated group if they were born before 1989 and had no history of vaccination at all. The proportion of EPI, non-EPI and unvaccinated students was 1533 (52.45%), 686 (23.47%) and 704 (24.08%) respectively (Table II).

Immunity to HBV (anti-HBs >/= 10 mIU/mL) was detected in 1314 (44.95%) of students made up of 519 (33.86%) of those in the EPI group, 596 (86.88%) of the non-EPI group and 199 (28.27%) of those not vaccinated (Table III). Hence, a large proportion (66.14%) of those vaccinated at birth had no detectable immunity by the time they reached the age for university admission. Among the 18 HBsAg positive students, 3 were in the EPI group, 4 in the non-EPI and 11 among the unvaccinated. In the EPI group, 6 of 1533 (0.39%) were found to have anti-HBc, including the 3 who were HBsAg positive. As anti-HBc is an indicator of HBV infection, the risk of infection among children vaccinated from birth was 0.39% compared to at least 0.58% (4 of 686) for the non-EPI group and 29.83% (210 [199+11] of 704) for the unvaccinated (results not shown in Table II). The risk of infection among the latter 2 groups was probably underestimated as these students were not tested for anti-HBc. Nonetheless, it is clear that students with HBV vaccinations (EPI or non-EPI) had lower risks of HBV infection compared to the non-vaccinated group (OR=0.012, 95% CI, 0.006-0.022).

Table I: Demographic data of students admitted to the Faculty of Medicine and Faculty of Dentistry, University of Malaya, 2005 to 2011

Year of intake	Course of study						Ethnic group				Gender	
	Bio-medical sciences	Nursing Nursing	Pharmacy	Medicine	Dentistry	Bio-imaging	Malay	Chinese	Indian	Others	M	F
2005	39	34	59	195	67		229	142	22	1	111	283
2006	49	34	61	196	78		234	161	19	4	129	289
2007	49	39	57	199	77		226	159	24	12	127	294
2008	34	34	65	200	77		204	177	22	7	126	284
2009	37	31	72	198	69		222	167	11	7	127	280
2010	36	33	70	196	92	18*	258	164	19	4	126	319
2011	35	29	72	200	69	23	238	176	12	2	123	305
Total	279	234	456	1384	529	41	1611	1146	129	37	869	2054

* Program started in 2010

Table II: Hepatitis B vaccination history and seroprevalence of HBsAg, Anti-HBc by age group

Age group	Total (%)	History of HBV vaccination				
		Vaccinated (%)		No History of vaccination (%)	HBsAg reactive (%)	Anti-HBc (%)
		EPI	Non-EPI			
Born ≤1958	3 (0.10)	0	2(66.67)	1 (33.33)	0	ND
1959-1968	42 (1.44)	0	33 (78.57)	9 (21.43)	1 (2.38)	ND
1969-1978	159 (5.44)	0	139 (87.42)	20 (12.58)	3 (1.89)	ND
1979-1988	1186 (40.57)	0	512 (43.17)	674 (56.83)	11 (0.93)	ND
Born ≥1989	1533(52.45)	1533 (100)	0	0	3 (0.20)	6 (0.39)
Total	2923	1533	686	704	18	6

HBV, hepatitis B virus; EPI, Expanded programme on Immunization; HBsAg, hepatitis B virus surface antigen; ND, not done

Table III: Anti-HBs and history of HBV vaccination

History of vaccination		Anti-HBS detected (%)	Anti-HBS not detected (%)	Total (%)
Vaccinated	Non-EPI	596 (86.88)	90 (13.12)	686 (23.47)
	EPI	519 (33.86)	1014 (66.14)	1533 (52.45)
Not vaccinated		199 (28.27)	505 (71.73)	704 (24.08)
Total		1314 (44.95)	1609 (55.05)	2923 (100.00)

EPI, Expanded Programme on Immunization; anti-HBs, antibody to the hepatitis B virus surface antigen

DISCUSSION

The EPI was established in 1974 through a World Health Assembly resolution to ensure that all children in all countries benefited from life-saving vaccines [10]. Following the introduction of universal infant HBV immunization in 1992[11], the WHO Western Pacific Region (WPR) adopted in 2005, a regional goal of hepatitis B control to reduce the prevalence of chronic HBV infection as indicated by the seroprevalence of HBsAg to less than 2% by 2012 and ultimately to less than 1% in children 5 years of age and below. In many countries in the WPR, the seroprevalence of HBsAg is still over 8% but the importance of universal vaccination is evidenced by the dramatic reduction in HBsAg seroprevalence among children in countries that have implemented nationwide infant vaccination programmes [12].

In this study on new students entering a Malaysian university for medical and paramedical studies, the HBsAg seroprevalence declined from 1.27% in 2005 to 0% in 2011, in line with increasing proportions of students vaccinated under the EPI. This trend attested to the efficacy of the universal infant HBV vaccination programme for protection against chronic HBV infection, for at least 18-19 years, without a booster vaccination. This protection, however, was not complete as there was still a 0.20% risk of infection among those vaccinated from birth. These break-through infections might be caused by high maternal viral load, inadequate immune response to the first course of vaccination, diminished immunity afterwards, or infection with vaccination escape mutants (VEMs). It has been reported that VEMs selected out by anti-viral treatment have been transmitted to individuals immunized with the HBV vaccine and that HBV immunization for infants might not prevent transmission of VEMs from mothers [13]. As the use of antivirals for the treatment of HBV infection increases, VEM-related infections and consequently vaccine breakthrough infections would become increasingly common [14].

Anti-HBs was not detected in 66.14% of those vaccinated at birth compared to only 13.12% of those vaccinated voluntarily at an older age (Table III). This difference is most likely to be due to the longer time interval between vaccination and testing in the first group. The duration of effective protection after infant vaccination and the need as well as timing for a booster vaccination has been long debated. North American and European authorities do not recommend booster vaccinations for immunocompetent individuals who have responded to a primary course of vaccination [15,16]. This advice is based on the observation that immunological memory seems to last for at least 15 - 18 years, in adequately vaccinated immunocompetent individuals, in the form of persisting anti-HBs and/or in vitro B-cell stimulation or an anamnestic response to a vaccine challenge [16]. Nevertheless, researchers have found that the geometric titres of vaccine-induced anti-HBs decline with time [17] with the persistence of protective antibody strongly

predicted by the extent of the maximal antibody response to vaccination and the long-term risk of HBV infection inversely related to the maximal antibody response to vaccination [18]. In Malaysia, as in many other countries, the adequacy of this antibody response is mostly assumed as there is no routine testing for seroconversion following vaccination. In Taiwan, Lin *et al.* [19] reported 37.4% anti-HBs prevalence among children 12 years after full primary neonatal vaccination, and Lu *et al.* [20] found HBV infection in 1/3 of children 15 years after initial vaccination. In the Netherlands, del Canho *et al.*[21] reported a child with an adequate antibody response (about 60 mIU/mL of anti-HBs at the age of one year) becoming HBsAg reactive in the fifth year of follow-up. Among children in Alaska, Peterson *et al.* [22] found that 88% of those vaccinated at birth had no detectable anti-HBs at the age of 5 years and one third failed to demonstrate an anamnestic response to a booster dose. These reports indicated a decline of protective antibody to undetectable levels in a substantial proportion of children less than 15 years after primary vaccination in infancy and, more importantly, a not infrequent failure of immunological memory on revaccination. Hence, some countries currently still have a policy of administering booster doses to certain risk groups, to provide reassurance of protective immunity against breakthrough infection. One of these risk groups consists of healthcare workers for whom HBV infection is an important occupational hazard. Tan *et al.* [23] reported an infection rate of 25% in about 2500 health employees in the state of Negri Sembilan, Malaysia, among whom the HBsAg was positive in 2.1% and anti-HBs in 22.8%, with increased relative risks among medical assistants and laboratory staff. It is a common practice, in most parts of Malaysia, to give a booster vaccination to those entering a medical or other healthcare profession. There is evidence that the anamnestic response elicited by a booster vaccination might not be able to re-establish protective memory within a week [24]. This delayed response might cause infection in some of those exposed to highly infectious patients. Hence, there may still be a place for regular boosters and follow-up monitoring for at- risk healthcare workers.

CONCLUSION

Overall, there is serological evidence that the universal infant HBV immunization program in Malaysia has been effective in reducing the rate of chronic HBV infection in children up to about 19 years of age. However, further investigations are necessary to evaluate the adequacy of the current vaccination programme for special at-risk groups, as well as the need for booster doses and long term monitoring, for early detection of clinically significant breakthrough infection or the carrier state. A catch-up HBV vaccination program is needed to vaccinate the students born before the implementation of EPI who have no immunity to HBV infection.

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REFERENCES

1. Custer B, Sullivan SD, Hazlet TK, Iloeje U, Veenstra DL, Kowdley KV. Global epidemiology of hepatitis B virus. J Clin Gastroenterol. 2004; 38(10): S158-68.
2. Hepatitis B vaccines: WHO position paper, Wkly Epidemiol Rec 2009; 84: 405–20 http://www.who.int/wer/2009/wer8440/en/index.html.
3. World Health Organization. Statistics on hepatitis B. Geneva: WHO, 2007.
4. Chen HL, Chang MW, Ni YH, *et al.* Seroepidemiology of hepatitis B virus infection in children - ten years of mass vaccination in Taiwan. JAMA, 1996; 276: 906-8.
5. Ni YH, Chang MH, Huang LM, *et al.* Hepatitis B virus infection in children and adolescents in a hyperendemic area: 15 years after mass hepatitis B vaccination. Ann Intern Med. 2001; 135: 796-800.
6. Poovorawan Y, Theamboonlers A, Vimolket T, *et al.* Impact of hepatitis B immunization as part of EPI. Vaccine, 2001; 19: 943-9
7. Lopez CG. Epidemiology of persistent hepatitis B virus infection. Malaysian J Pathol 1985; 7: 7-10.
8. Ng KP, Saw TL, Baki A, Rozainah K, Pang KW, Ramanathan M. Impact of Expanded Programme of Immunization on hepatitis B infection in school children in Malaysia. Med Microbiol Immunol 2005; 194: 163-8.
9. Committee of Vice-Chancellors and Principals of the Universities of the United Kingdom. Guidance on fitness to practise: hepatitis B. London: The Committee, 1995.
10. World Health Assembly (27th:1974). The Expanded Programme on Immunization: the 1974 resolution by the World Health Assembly. Assignment Child 1985; 69-72: 87-8.
11. World Health Organization. Expanded programme on immunization. Global advisory group – part I. Wkly Epidemiol Rec 1992; 67: 11-5.
12. Rani M, Yang B, Nesbit R. Hepatitis B control by 2012 in the WHO Western Pacific Region: rationale and implications. Bull World Health Organ 2009; 87: 707-13.
13. Hwang EW, Cheung R. Global epidemiology of hepatitis B virus (HBV) infection. North Am J Med Sc 2011; 4(1): 7-13.
14. Thibault V, Aubron-Olivier C, Agut H, Katlama C. Primary infection with a lamivudine-resistant hepatitis B virus. AIDS 2002; 16: 131-3.
15. European Consensus Group on Hepatitis B Immunity. Are booster immunisations needed for lifelong hepatitis B immunity? Lancet 2000; 355: 561-5.
16. MMWR Recommendations and Reports. Hepatitis B virus: A comprehensive strategy for eliminating transmission in the United States through universal childhood vaccination. Recommendations of the Immunization Practices Advisory Committee (ACIP). MMWR Recomm Rep 1991; 401: 1-25.
17. Huang LM, Chaing BL, Lee CY, Lee PI, Chi WK, Chang MH. Long-term response to hepatitis B vaccination and response to booster in children born to mothers with hepatitis e antigen. Hepatology 1999; 29: 957-9.
18. Fujisawa T, Onouse M, Inui A, Kosugi T. Serial changes in titters of antibody to hepatitis B surface antigen after immunzatiuon of infants born to mothers with hepatitis B e antigen. J Pediatr Gastroenterol Nutr 1996; 23: 370-4.
19. Lin YC, Chang MH, Ni YH, Hsu HY, Chen DS. Long-term immunogenicity and efficacy of universal hepatitis B virus vaccination in Taiwan. J Infect Dis 2003; 187: 134–8.
20. Lu CY, Chiang BL, Chi WK *et al.* Waning immunity to plasma-derived hepatitis B vaccine and the need for boosters 15 years after neonatal vaccination. Hepatology 2004; 40: 1415–20.
21. del Canho R, Grosheide PM, Mazel JA, et al. Ten-year neonatal hepatitis B vaccination program, The Netherlands, 1982-1992: Prospective efficacy and long-term immunogenicity. Vaccine 1997; 15: 1624-30.
22. Petersen KM, Bulkow LR, McMahon BJ, *et al.* Duration of hepatitis B immunity in low risk children receiving hepatitis B vaccination from birth. Pediatr Infect Dis J 2004; 23: 650-5.
23. Tan TC, Vadivale M, Ong CN. Prevalence of Hepatitis B Surface Antigen and Antibody among Health Care Employees in Negri Sembilan, Malaysia, 1989. Asia Pac J Public Health 1992-1993; 6: 134-9.
24. Yishida T, Saito I. Hepatitis B booster vaccination for healthcare workers. Lancet 2000; 355(9213): 1464.