ORIGINAL ARTICLE

Immunogenicity of Low Dose Recombinant DNA Hepatitis B Vaccine in Children Ten Years Old and Younger

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Abstract To evaluate the usefulness of low dose hepatitis-B vaccine, we recruited 70 healthy children of both sexes aged ten years or less. To be included in the study a child must have been clinically healthy and gave negative results for hepatitis-B virus infection (HBsAg, anti-HBs, anti-HBc), and with seerum glutamic oxaloacetic acid transaminase less than 50 mU/l. Subjects with intercurrent infection and those with chronic disease or moderate to severe malnutrition were excluded from the study. They were given 3 injections of 2.5 ml rDNA hepatitis B vaccine intramuscularly on month 0, month 1, and month 6. The seroconversion rate after the third injection in all subjects was 92.3%; this was slightly different between the well-nourished and under-nourished children (93.9% vs. 87.5%), and between girls and boys (96.7% vs 88.6%). The only side effect noted was fever encountered in 1 subject. We conclude that three intramuscular injections of 2.5 mg of rDNA hepatitis B vaccine give good anti-HBs titer, so that this method may be considered to be used in the mass immunization program. **[Paediatr Indones 1997; 37:76-85]**

Introduction

Indonesia is one of the regions which has the highest prevalence of hepatitis B virus (HBV) infection.[1,2] In children <14 years old the HBV infection prevalence is 9.17%, and it is even higher in East Nusa Tenggara/Lombok Island as it reaches 18.8% in children aged 14 years.[1] The HBV carrier rates in pregnant women is 5%.[2] Considering

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the seriousness of the problem, efforts to control the disease are mandatory to prevent the harmful effect of HBV infection.[3-6]

The income of most Indonesian people is too low to afford the self-supporting vaccination. A lower cost of hepatitis B vaccination will certainly be an answer for increasing the coverage of the self-supporting vaccination program.[7,8] This study aimed to evaluate the immunogenicity and the efficacy of 2.5 mcg (half of previously recommended dose) H-B-VAX® II in children ≤ 10 years old.

Methods

Subjects

Healthy children of ≤ 10 years old, living in "Pondok Timur Mas" residential estate, Jaka Setia-Bekasi, West Java, were included in the study. Subjects with intercurrent infections, those having suffered from chronic diseases and moderate to severe malnutrition, were excluded from the study. With initial examination of 100 healthy children, it was expected to have 50-75% (n=60) children with negative serological findings for HBV infection, i.e., HBSAg, anti-HBs, and anti-HBc; with SGOT < 50 mU/l. Informed consent were obtained from the parents. Each child was weighed by experienced nurse. The data were recorded and the nutritional status of the subjects were classified according to the Waterlow classification.

Study Design

This descriptive study was carried out over an 8 months period. There were 100 "healthy" children who were screened for HBsAg, anti-HBs, anti-HBc, and SGOT. All eligible subjects, 70 healthy children, were included in this study. They were given 3 doses of 2.5 mg H-B-VAXRII (MSD recombinant DNA hepatitis B vaccine). All doses were given at time 0,1 and 6 months, intramuscularly, at the anterolateral thigh.

Results

Immunogenicity

Immunogenicity as related to age

Table 1 shows the anti-HBs responses in all the subjects ranged 10 months to 10 years old. The result after the second injection revealed that 40 subjects (61.5%) had

seroconversion and 25 subjects (38.5%) showed no seroconversion (negative anti-HBs). After the third injection, 60 subjects (92.3%) have had seroconversion and only 5 children (7,7%) remained anti-HBs negative. The GMTs after second and third injections were 55 and 5289 mIU/ml.

Table 1. Seroconversion rates and GMTs of anti-HBs in children 0-10 years

Injection	Anti-HBs responses				
	Positive			Negative	
	N	%	GMT (mIU/ml)	N	%
Second	40	61.5	55	25	38.5
Third	60	92.3	5,289	5	7.7

The immune responses among children < 5 years is shown in Table 2. After the second injection it showed that 65.4% of these children have had seroconversion with the GMT of 78 mIU/ml. After the third injection 96.2% of children < 5 years old have had seroconversion and the GMT was 3,414 mIU/ml.

Table 2. Seroconversion rates and GMTs of anti-HBs in children < 5 years

Injection	Anti-HBs responses				
	Positive			Negative	
	N	%	GMT (mIU/ml)	N	%
Second	17	65.4	78	9	34.6
Third	25	96.2	3,414	1	3.8

Immunogenicity as related to sex

The responses among each sex group are shown in Tables 3 and 4. The male subjects had seroconversion rates of 57.1% and 88.6% after the second and third injections, respectively. The seroconversion rates in girls were 66.7% and 96.7% after second and third injections, respectively. The GMTs after the second and third injections were 52 and 7555 in girls, and 57 and 3789 in boys.

Table 3. Seroconversion rates and GMTs of anti-HBs in boys

Injection	Anti-HBs responses				
	Positive			Negative	
	N	%	GMT (mIU/ml)	N	%
Second	20	57.1	57	15	42.9
Third	31	88.6	3,789	4	11.4

Table 4. Seroconversion rates and GMTs of anti-HBs in girls

Injection	Anti-HBs responses				
	Positive			Negative	
	N	%	GMT (mIU/ml)	N	%
Second	20	66.7	52	10	33.3
Third	29	96.7	7,555	1	3.3

Immunogenicity as related to the nutritional status

The anti-HBs responses among each nutritional status group are shown in Tables 5 and 6. The well nourished subjects had seroconversion rates of 57.1% and 93.9% after the second and third injections with the GMTs of 55 and 5396 mIU/ml. The seroconversion rates in mildly undernourished subjects were 75.0% and 87.5% after the second and third injections. The GMTs after the second and third injections in mildly undernourished subjects were 52 and 4952 respectively.

Table 5. Seroconversion rates and GMTs of anti-HBs in well-nourished group

Injection	Anti-HBs responses				
	Positive			Negative	
	N	%	GMT (mIU/ml)	N	%
Second	28	57.1	55	21	42.9
Third	46	93.9	5,396	3	6.1

Table 6. Serocoversion rates and GMTs of anti-HBs in under-nourished group

Injection	Anti-HBs responses				
	Positive			Negative	
	N	%	GMT (mIU/ml)	N	%
Second	12	75.0	52	4	25.0
Third	14	87.5	4,952	2	12.5

Quantitation of anti-HBs

After the second injection, the antibody levels in 40 seroconverter subjects ranged from 8 mIU/ml-945 mIU/ml. About 29 (72,5%) subjects had antibody levels of 10-100 mIU/ml and 10 (25%) subjects had > 100 mIU/ml (Table 7). Only one subject (2.5%) had the very low level of anti-HBs (8 mIU/ml), which was considered to be not protective.

The antibody levels after the third injection in the 60 seroconverters were 28-109 000 mIU/ml. No subjects developed antibody levels of less than 10 mIU/ml. About 7 (11.7%) subjects had the levels of 10-100 mIU/ml, 6 (10%) had antibody levels of 101-1000 mIU/ml, and 47 (78,3%) subjects with > 1000 mIU/ml (Table VII).

Table 7. Levels of antibody after second and third injection

Injection	Titer of anti-HBs (mIU/ml)			
	< 10	10-100	101-1000	> 1000
Second	1 (2.5%)	29 (72.5%)	10 (25%)	0
Third	0	7 (11.7%)	6 (10%)	47 (78.3%)

Anti-HBs levels as related to sex

The antibody levels in both sexes are shown in Figure 1. After the second injection, 14% of the male subjects had antibody levels of > 100 mIU/ml and 43% had antibody of 10-100 mIU/ml; while 43% were anti-HBs negative. After the third injection, 66% boys had antibody of > 1000 mIU/ml, 9% had antibody levels of 101-1000 mIU/ml, and 14% had 10-100 mIU/ml; about 11% did not develop antibody (Figure 1).

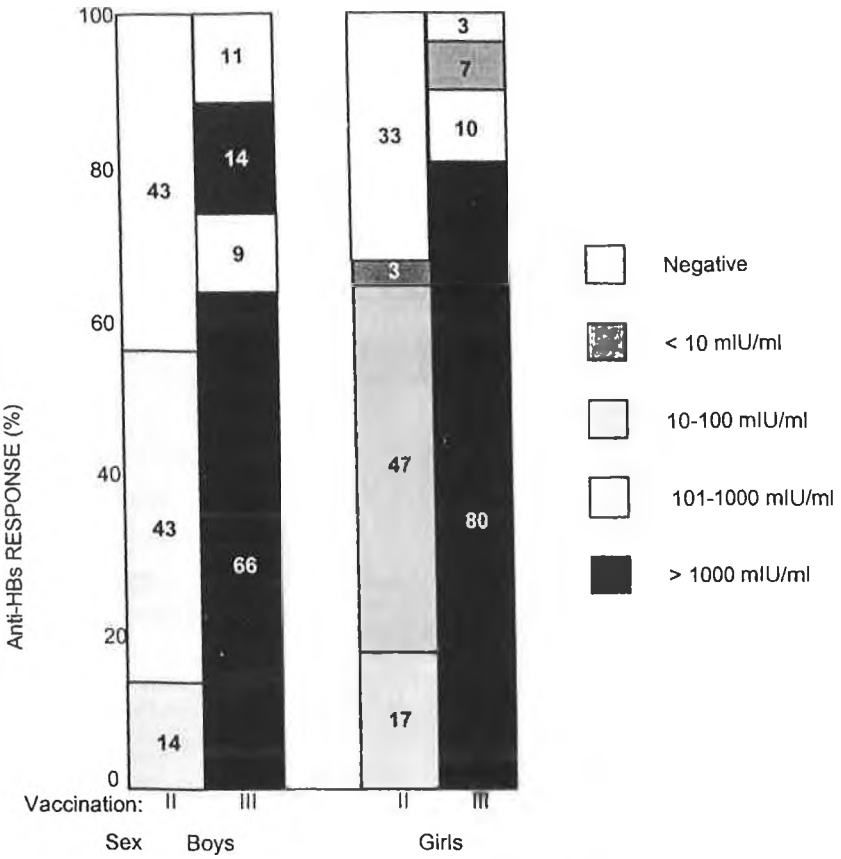

Figure 1. Anti-HBs levels by sex

In female subjects, the antibody levels after the second injection in 17% girls were > 100 mIU/ml, in 47% were 10-100 mIU/ml, in 3% were < 10 mIU/ml; and 33% were anti-HBs negative. After the third injection, 80% of the girls had antibody levels of more than 1000 mIU/ml, 10% had antibody levels ranged 101-1000 mIU/ml, and 7% with the levels ranged 10-100 mIU/ml, and about 3% were anti-HBs negative (see Figure 1). Neither male nor female vaccinees had antibody levels < 10 mIU/ml after the third injection.

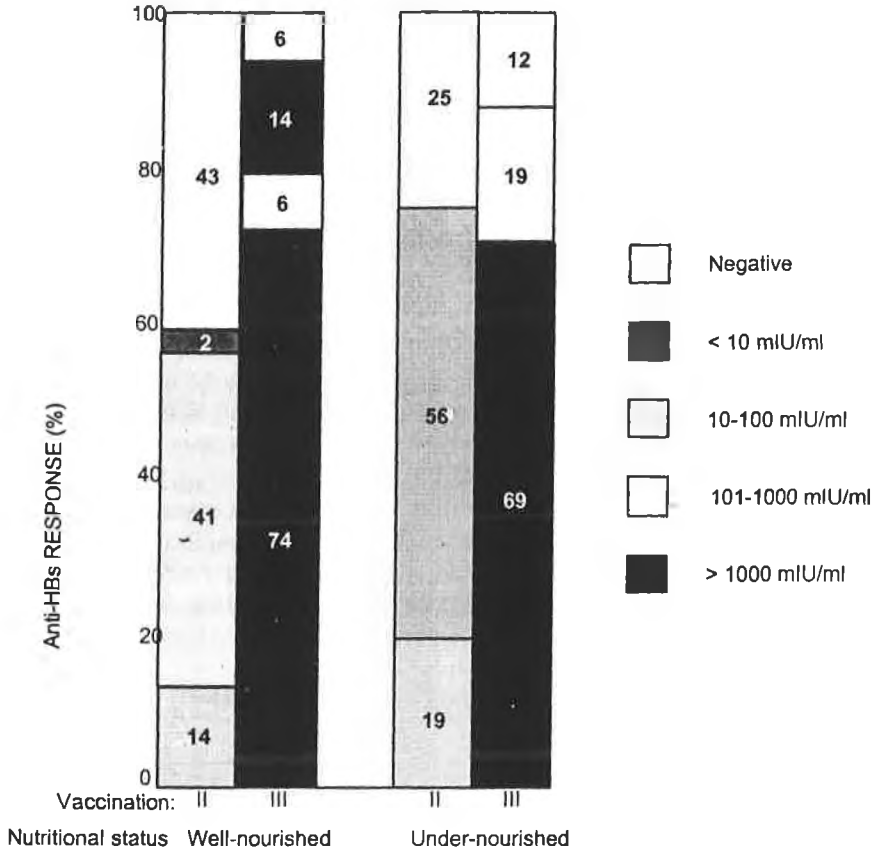

Figure 2. Anti-HBs levels by the nutritional status

Anti-HBs level as related to the nutritional status

After the second injection 14% of well-nourished-subjects (see Fig. 2) had antibody levels of > 100 mIU/ml, 41% with the level of 10-100 mIU/ml, 2% with the levels of < 10 mIU/ml, and 43% subjects did not develop antibody. After the third injection, 74% had antibody levels of > 1000 mIU/ml, 6% had 101-1000 mIU/ml, 14% had 10-100

mIU/ml, no subjects had antibody levels of <10 mIU/ml, while 6% subjects were anti-HBs negative.

In mildly undernourished subjects, after the second injection 19% subjects had antibody levels > 100 mIU/ml, 56% had 10-100 mIU/ml, no subjects had the levels of < 10 mIU/ml and about 25% subjects did not develop antibody. After the third injection, 69% subjects had antibody levels > 1000 mIU/ml, 19% subjects had 101-1000 mIU/ml antibody levels, no subjects with antibody levels of <100 mIU/ml and only 12% subjects with no detectable antibody (Figure 2)

Table 8. Clinical complaints during a-5-day period after vaccinations

Type of complaints	First injection		Second injection		Third injection	
	Yes	No	Yes	No	Yes	No
Fever	1	64	1	64	0	65
Injection site						
Redness	0	65	0	65	0	65
Soreness	0	65	0	65	0	65
Systemic*	0	65	0	65	0	65

* Nausea, headache, diminished appetite, cold symptoms

Discussion

Hepatitis B virus infection is endemic in Indonesia and the carrier states are about 5%-19%.[1] Infection during childhood forms the main bulk of the carrier pool. Vaccination is the only effective means of preventing HBV infection and its sequelae of chronic liver disease and/or hepatocellular carcinoma.[3,5,9] Therefore, HBV infection is a major public health problem in Indonesia as in other developing countries.

The cost of recombinant DNA (rDNA) hepatitis B virus vaccine varies in different countries. Previous studies revealed that vaccination with three doses of 2.5 μg, have been shown to be highly immunogenic in young children.[4,8] Economic advantage will surely be gained from this low dose of regimen.[3,8,9]

The present study revealed very high seroconversion rates and reasonable GMT levels of children given 2.5 μg dose of three dose vaccines. Antibody responses for the MSD rDNA hepatitis B vaccine was 92.3% after the third injection, nearly similar with other studies using the same dose. The seroconversion rates in subjects ≤ 5 years old were quite high (96%). Indicating that this reduced dose regimen did produced

satisfactory result, even in such young children. About 72% had antibody level of more than 1000 mIU/ml and the GMTs was increased nearly 100 times after the third injection. Even the low responder (1 sample) turned out to be having a very high antibody level after the third injection. The seroconversion rates, the antibody levels, and the increased of the GMTs, were higher in girls compared to the boys. Whether these results happened by chance or not, needs further evaluation.

Among the well-nourished-vaccines, the seroconversion rates, the percentage of high antibody levels, and the increased of the GMTs, were higher than the undernourished-vaccinees. These findings are understandable since well-nourished children have better immune response than children of less well-nourished.

The duration of vaccine-induced immunity was not evaluated in this study. According to literature, in about 20-60% of adult vaccines the anti-HBs levels decreased to less than 10 mIU/ml, 5-9 years after immunization. Yet, after an exposure with HBV the immunologic memory was retained, and anamnestic antibody responses occurred after exposure to HBV.

Conclusions

Antibody responses of 2.5 μg MSD rDNA hepatitis B vaccine (HB-Vax II) in children 10 years old or younger were proved to be high with a seroconversion rate of 92.3%. Regarding the efficacy and affordable cost of the 2.5 μg doses of the MSD recombinant product, this low dose regimen could be considered for mass immunization program, toward the important target of children 10 years old or younger. This recombinant hepatitis B vaccine is also well tolerated, and no serious adverse reactions attributable to vaccine have been reported. The relationship between antibody persistence and duration of protection need to be fully defined by doing long-term clinical studies.

Acknowledgments

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