The Comparison Of Different Recombinant Hepatitis-B Vaccines

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Abstract

Objectives: To compare safety and immunogenicity of four recombinant hepatitis B (HB) vaccines in Turkish market in different healthy ages groups.
Methods: Overall, 265 subjects were administered with one of the four different HB vaccines at 0, 1, 6 months according to prescribing information and under routine hospital practice.
Results: Seroprotection rates were similar between the vaccination groups approximately 7-8 months after administration of the initial dose (98%, 98%, 97% and 100%, p<0.05, respectively). The mean titres at 6 months were 1386 (2-27.400) mIU ml-1 in group I, 1422 (6-19.700) mIU ml-1 in group II, 1448 mIU ml-1 (3-29.300) mIU ml-1 in group III, and 1497 (110-19.800) mIU ml-1 in group IV, respectively.
Conclusions: All of the recombinant vaccines in the Turkish market were found effective in this study.

INTRODUCTION

Hepatitis B is a disease of major importance with more than 2 billion people being infected globally with the hepatitis B virus (HBV), with approximately 350 million as term carriers of HBV [1]. Hepatitis B surface antigen (HBsAg) positivity is still an endemic problem in Turkey with between 5 and 10% of the population testing positive for the marker of infection [2]. These chronically infected individuals have significantly increased risk of developing many long - term sequelae, including chronic active hepatitis, cirrhosis of the liver, and primary hepatocellular carcinoma [3].

No current medical treatment is totally effective in the treatment of chronic HBV infection [4]. Thus, carriers will continue to serve as sources of new infections for susceptible persons (5). The most effective means of controlling the spread of HBV is prophylactic or postexposure immunization. Prevention of hepatitis B by active immunization became a reality more than two decade ago, with the availability of plasma-derived, and subsequently recombinant DNA, hepatitis B vaccines (6, 7). Current recommendation by the Centers for Disease Control and Prevention (CDC) include vaccination of emerging at – risk populations, such as infants, adolescents and susceptible contacts of chronic HBV carriers; high risk individuals such as intravenous drug users; and persons with occupational risk (i.e. any health – care or public – safety worker); and individuals postexposure (8).

Since 1990s, hepatitis B vaccine has been integrated in to the current childhood and adult immunization schedules on a nationwide basis. Vaccination is now advisable for newborn and for children entering puberty (at about 12 years old) and high risk group immunization is expected to be intensified (9). Currently, four hepatitis B vaccines are being marketed in Turkey. They are: (1) Gen Hevac B (Pasteur, Merieux, France), (2) Engerix – B (Hepatitis B vaccine, recombinant, Smith Kline Beecham, Philadelphia, PA), (3) Hepavax Gene (Korea Green Cross Co., Seoul, South Korea) and (4) HB Vax II (Merck Sharp & Dohme). All of them are yeast derived and produced by genetic engineering methods. The vaccination schedule most often used in adults and children is a series of theree intramuscular (IM) injections, the second and third dose administered 1 and 6 months, respectively, after the first. This recommended series induces a protective antibody response (anti-HBs =100 international units IU ml $^{-1}$) in 90% of healthy adults and in 95% of infants, children, and adolescents (10, 11).

The purposes of this vaccine evaluation study were a) to compare the immunogenicity and safety of four recombinant DNA vaccines, licenced in Turkey, and b) to determine the

immunogenicity among individuals randomly assigned based on selected age groups. The vaccines are Engerix B and Hepavax Gene, which contain only the S polypeptide, and Gen Hevac B and HB Vax II, which contains both the PreS1 and PreS2 proteins, as well as the S antigen.

MATERIALS AND METHODS

VACCINES

Gen Hevac B, Engerix – B, Hepavax Gene, and HB Vax II were used in this study. The HB Vax II, Engerix – B and Hepavax Gene vaccines contain S protein and are produced in yeast (Saccharomyces cc revision). In contrast, the Gen Hevac B vaccine contains PreS1, PreS2 and the S protein and is produced by Chinese hamster ovarian cells. Each 1-mL vaccine dose contained 20 µg and 0.5 mL vaccine dose contained 10 µg of hepatitis B surface antigen (HBsAg), adsorbed on aluminium hydroxide as adjuvant, and thimerosal as a preservative. Each 1-ml vaccine dose of HB Vax II contained 10 µg and 0,5 ml vaccine dose contained 5 µg of HBsAg.

STUDY POPULATION AND DESIGN

This was a prospective vaccine evaluation study, under routine practice in military hospital with healthy population without age limits. A total of 265 healthy personnel assigned to our hospital were enrolled and 112 of those were introduced to Gen Hevac B, 80 to Engerix-B, 36 to Hepavax Gene and 37 to HB Vax II. All recipients of the vaccine underwent screening for HbsAg, hepatitis B core antibody (anti-HBc total) and hepatitis B surface antibody (anti-HBs) before the first vaccination. Only anti-HBc total and anti-HBs-negative subjects were recruited. Volunteers were excluded if they had previously been vaccinated with HBV vaccine or had a prevaccination serum specimen that was positive for HBsAg or anti-HBs and had previously demonstrated hypersensitivity reaction to yeast; had significant and persistent hematologic, hepatic, renal, cardiac, or respiratory disease; were concurrently enrolled in another clinical trial; were receiving immunosuppressive therapy, or had received immunoglobulin, blood or blood products within the previous 180 days, or a non-FDA approved drug within the 30 days prior to the study initation. Female volunteers who were pregnant or lactating were also excluded from study participation. All volunteers provided written informed consent prior to trial initiation. Institutional review board approval for the protocol was obtained at each investigational site prior to study initiation.

Vaccines were administered intramuscularly in the deltoid muscle at 0, 1, and 6 months. When the first dose was administered at month 0, appointments were made for the second and third vaccinations. The month 1 time point for the second vaccination ranged from 26 to 34 days after the first dose, and the month 6 time point for the third dose was proposed as 5 to 7 months after the first vaccination. The time intervals for vaccine administation at months 1 (± 4 days) and 6 (± 1 month) were decided for reasons of practicality and flexibility to enable a maximum number of vaccines to complete the immunization course.

Before the vaccination, a complete medical history and physical examination were performed in all volunteers and blood was drawn for determination of HBsAg and anti-HBs. For those deemed eligible to participate, the first dose of the vaccine was administered. Each volunteer was observed for 15 min after vaccination for any adverse events, and asked to report verbally the occurrence and severity of any local or general symptoms (fever, nausea, or malaise) on the day of vaccination and for the following 2 days to the pysicians who performed the immunizations. During the approximate 7-months period of observation for each vaccinated person, any serious adverse event was to be reported.

LABORATORY ANALYSIS

Anti-HBs titers were to be measured 4 to 6 weeks after the third vaccine dose according to the routine practice of the hospitals, by using a range of an enzyme-linked immunosorbent assay. Antibody titers were measured by using standardized test kits that are licensed and commercially available in Turkey (Bioelisa AntiHBsAg, Biokit, Barcelona, Spain) at the Microbiology laboratory of Kasimpasa Nawal Hospital. Titers of anti-HBs were expressed as mili-international units per mililiter (mIU ml $^{-1}$). A titre lower than 10 mIU ml $^{-1}$ was defined as a failed response to vaccination. A titre of =10 to =99 mIU ml $^{-1}$ was defined as a partial response. Titres =100 mIU ml $^{-1}$ were defined as a good response to the vaccination ($_{12}$). The seroprotection rate (SPR [given as a percentage]) was the proportion of vaccinces with an anti-HBs titer of 100 IU mIU ml $^{-1}$ or greater, which was the minimum titer that was considered to be protective as defined by the World Health Organization (^{12}). As a control group, 15 healthy personnels who have had history of hepatitis B virus infection, had anti-HBc and anti-HBs that was over 100 mIU ml $^{-1}$, were selected. Anti-HBs test was standardized by measuring the values of GMC in 15 controls. The values were compared to different anti-HBs ELISA kits (International Immunodiagnostics, Carson City, Nevada-USA).

STATISTICAL ANALYSIS

In the exploratory statistical analysis that was performed the gross seroresponse rates were compared by using a x^2 test, and the GMTs were compared by using an unpaired t tests. Comparison of GMTs was done by using unpaired T tests or F tests. A pragmatic approach was used for ranking the explanatory variables according to their influence on the target variables by applying the Fisher combination of P values on those derived for the individual tests for GMTs and gross response rates. For all statistical tests, the 2-sided P values were calculated at the 5% significance level. Statistical analyses were performed by using a commercially available software package (SAS Institute, Cary, NC). A P-value = 0.05 (two-tailed) was considered to be significant.

RESULTS

A total of 265 heathy persons were eligible for participation and received three doses of the vaccine. Five volunteers in group I, seven volunteers in group II, and one volunteer in group IV did not complete the study or had no evaulable immunogenicity data and were not included in the final analysis. Table I summarizes the demographic parameters of volunteers with evaluable immunogenicity data. Evaluable volunteers in four vaccination groups were demographically comparable for age and gender. There were no differences between any of the groups in terms of age, gender or later identifiable factors.

Figure 1

TABLE I. Demographic parameters for evaluable volunteers by vaccination group

Parameter	Gen Hevac-B (n=112)(%)	Engerix-B (n=80)(%)	Hepavax Gene (n=36)(%)	HB Vax II (n=37)(%)
Gender				
Male	69	46	21	23
Female	43	34	15	14
Mean age (±SD.)	29.5±7.8	28.3± 6.8	30.2±7.4	27.9±5.4
Age disturbance	3 (3)	4 (5)	-	-
0-10	12 (11)	10 (13)	4 (11)	5 (14)
11-20	31 (28)	35 (44)	16 (44)	13 (35)
21-30	30 (27)	22 (28)	12 (33)	13 (35)
31-40	26 (23)	6 (8)	4 (11)	5 (14)
41-50	4 (4)	1 (1)	-	1 (3)
51-60	**1 (1)**	-	-	-
61-70	**5 (4)**	2 (3)	-	-
71-80	**112**	80	36	37
TOTAL				

IMMUNOGENICITY

Overall, the SPR was found as 98% (259/265). The SPR was 98% (110/112) in group I, 98% (78/80) in group II, 97% (36/37) in group III and 100% (37/37) in group IV, respectively. The anti-HBs GMT at 6 months were 1386 (2-27.400) mIU ml^{-1} in group I, 1422 (6-19.700) mIU ml^{-1} in group II, 1448 (3-29.300) mIU ml^{-1} in group III and 1497 (110-19.800) mIU ml^{-1} in group IV (Table II). Five subjects who did not respond in this study were older with a greater proportion of males (60% vs 64%) when compared to the remaining subjects in whom anti-HBs titers (=10 mIU mL^{-1}) had developed (40.2 vs 32.4 years).

When stratified by sex, the SPRs were similar among males, 98% (156/159) and females, 98% (104/106). However, males were associated with a lower GMT (1225 mIU ml^{-1}) than were females (1726 mIU ml^{-1} , P=.04). In group I and II, no difference was found between males and females [(68/69) 99% vs. (42/43 98% and (45/46 (95%) vs (33/34) 97%]. However, the SPRs were greater, 100% (21/21) in males than in females, 93% (14/15) in group III (p= .06). The immunogenicity results, when classified by group, are

given in Table 2.

Figure 2

TABLE II. The Seroprotection Rate (SPR) and Geometric Mean Titers (GMT) in Vaccines groups

Parameters	Gen Hevac-B (n=112)(%)	Engerix-B (n=80)(%)
GMT (mIU/ml)	1386(2-27.400)	1422 (6-19.700)
Respond n (%)		
≥100 mIU ml^{-1}	85/112 (76)	66/80 (83)
≥10 mIU ml^{-1}	25/112 (22)	12/80 (15)
<10 mIU ml^{-1}	2/112 (2)	2/80 (3)
Respond (≥10 $mIUml^{-1}$) and Age n (%)		
0-10	3/3 (100)	4/4 (100)
11-20	12/12 (100)	10/10 (100)
21-30	30/31 (97)	35/35 (100)
31-40	30/30 (100)	21/22 (95)
41-50	25/26 (96)	5/6 (83)
51-60	4/4 (100)	1/1 (100)
61-70	1/1 (100)	-
71-80	5/5 (100)	2/2 (100)

Figure 3

Parameters	Hepavax Gene (n=36)(%)	HB Vax II (n=37)(%)	TOTAL (n=265)(%)
GMT (mIU/ml)	1448(3-29.300)	1497 (110-19.800)	
Respond n (%)			
≥100 mIU ml^{-1}	31/36 (86)	37/37(100)	219/265 (83)
≥10 mIU ml^{-1}	4/36 (11)	0/37(0)	41/265 (15)
<10 mIU ml^{-1}	1/36 (3)	0/37 (0)	5/265 (2)
Respond (≥10 $mIUml^{-1}$) and Age n (%)			
0-10	-	-	7/7 (100)
11-20	4/4 (100)	5/5 (100)	31/31 (100)
21-30	16/16 (100)	13/13 (100)	94/95 (99)
31-40	12/12 (100)	13/13 (100)	76/77 (99)
41-50	3/4 (75)	5/5 (100)	38/41 (93)
51-60	-	1/1 (100)	6/6 (100)
61-70	-	-	1/1 (100)
71-80	-	-	7/7 (100)

For subjects who completed with the 0-, 1-, and 6- month vaccination schedule, the SPR and GMT were 98% (110/112) and 1386 (2-27.400) mIU ml^{-1} (95% confidence interval [CI], 1169-1643 mIU ml^{-1}) in group I, 98% (78/80) and 1422 (6-19.700) mIU ml^{-1} (95% confidence interval [CI], 1201-1711 mIU ml^{-1}) in group II, 97% (35/36) and 1448 (3-29.300) mIU ml^{-1} (95% confidence interval [CI], 11237-1763 mIU ml^{-1}) in group III, and 100% (37/37) and 1497 (110-19.800) mIU ml^{-1} (95% confidence interval [CI], 1319-11827 mIU ml^{-1}) in group IV, respectively. There were no significant differences between the four groups for either the SPRs or GMTs.

The five subjects who did not respond in this study were 26 and 46 years old in group I, 33 and 49 years old in group II, and 42 years old in group III. By using the X^2 test, the SPRs were different in 41-50 years – old subjects, when compared to other groups. But this difference was not statistically significant ($p>0.05$). No serious adverse effects were seen in any subjects. There were no differences between groups with regard to identifiable adverse effects. The most common adverse effects were local pain (3.8 %), local redness at the injection site (2.3 %) and fever (0.8 %).

DISCUSSION

Today, people remain at high risk for the development of HBV infection in developing countries. Although HBV infection can be prevented with vaccination, not only does it remain an important health problem in countries where HBV is endemic, but also in countries where HBV is non – endemic. In vaccine clinical trials, administration of the standart regimen of 20 µg at 0, 1 and 6 months to healthy adults resulted in a SPRs of 96% at month 7 ([7]). Conversely about 10% of vaccinated individuals don't respond due to hepatitis B virus S gene variations and, as a result, remain at risk for the development of HBV infection ([1], [13]). In another study, the immunogenicity in over 40,000 healthy infants were evaluated and the SPR was found to be 98.6% ([14]). Thus, strategies to convert those non–responding individuals to anti-HBs responders have been of considerable importance, at least in endemic areas for HBV infection.

A different vaccine that contains both the Pre S1 and Pre S2 antigens, as well as the usual S protein, has been developed and has been reported to be more immunogenic. In one animal study, the Pre S1 and Pre S2 antigen – containing vaccine was effective in animals not previously responding to a standard S- antigen – only vaccine ([15]). In another study, a similar – response was reported in people who had not previously responded to a standard isolated S–antigen–containing vaccine. On the other hand, the Pre S2 antigen has been shown to be less immunogenic than the HBsAg ([16]). In our study, a standard isolated S-antigen–containing vaccine (5 and 10 µg) (Group IV) was more immunogenic than others when compared based on the GMT rates. But the SPR rates were similar with each groups.

The incidence of HBV infection increases rapidly during adolescence. Although the prevalence varries by region, gender and race; by 25-34 years of age, between 3.3% and

25% of all persons have had HBV infection ([16]). Although universal immunization of adolescents has the advantage of protecting these individuals during the "at risk" years, the vaccination of older age groups that have low immunity is problematic. Zajac and colleagues reported that antibody response declines with increasing age in recipients of recombinant DNA hepatitis B vaccine ([11]). The results from this study confirm that age plays a role on both the SPR and GMT, elicited after a three – dose schedule at 0, 1, and 6 months in group I, II, III. This finding has also been found in previous studies for SPRs, as well as for GMTs ([11]). However, the SPR rates was 100% and GMT rates was 1497 mIU ml $^{-1}$ in group IV, and no subjects older than 61 years were enrolled in group IV of the study.

Several different causes may be possible. First, the body mass index as a factor for a low anti-HBs response has also been found in several studies, although not specifically for GMTs ([17]). Second, it was reported that a higher yet nonsignificant number of vaccinees who smoked greater than 11 cigarettes per day had anti – HBs values less than 10 mIU/ml ([18]). Third, persons with chronic underlying diseases were significantly associated with lower GMTs than were vaccinees without chronic underlying diseases. For this reason, the SPR and GMT rates in group IV may have been greater than those observed from other studies.

Although most of study had reported short - term persistence of anti-HBs after hepatitis B vaccination, four papers have reported long - term persistence among health care workers. In a study, the persistence of anti – HBs in vaccinees 12 years after primary immunization and the response to a booster dose using a recombinant DNA yeast – derived hepatitis B vaccine was documented ([19]). In another study, a 85.4% seroprotection rate was observed after 6 years with a significantly higher seroprotective rate in subjects who received four doses of vaccines, including Pre S1 and S protein, during primary immunization when compared with to those who had received three doses of vaccines, including 5 µg S protein (93.9% versus 67.2% ([20]). After the booster dose, subjects who received vaccines, including Pre S1, Pre S2, and S protein during primary immunization were better seroprotected and had higher seroprotection rates.

The relationship between anti-HBs and protection from clinical infection has been demonstrated by many previous clinical studies conducted in the 1970s and 1980s. These studies demonstrated that high levles of anti-HBs, that is, 10 mIU/ml, protected vaccinees against hepatitis B infection ([21]). This was observed in chimpanzees, first with the plasma-derived vaccine in the 1970s and then with the yeast-derived vaccine in the 1980s ([22]). Subsequent clinical studies with both vaccines have demonstrated protective efficacy in high-risk human populations. Currently, yeast-derived recombinant hepatitis B vaccinees are common in practice under the condition of universal immunization and they are safe and well-tolerated, usually producing high immunogenicity ([11]). On the other hand, no serious adverse reactions, including immediate (anaphylaxis and urticaria) and delayed (skin, rheumatic vasculitis, hematologic, ophtalmologic and neurologic) autoimmune reactions were detected in our study ([23], [24]).

CONCLUSIONS

In conclusion, this prospective postmarketting study reinforces that the four different recombinant hepatitis B vaccines licenced in Turkey have a good tolerability and are highly immunogenic among all age groups.

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