Clinical trial of simultaneous administration of PRE S2 and S containing recombinant Hepatitis B vaccine and tetanus toxoid in nonresponder groups: A preliminary study

Emine SÖNMEZ[1], Halil İbrahim ÖZEROL[2], Yasemin ÇINARA, Kazım ŞAHİN[2], Hatice ÖZBİLGE[3], Gönül ARSLAN[3], Şerafettin YILMAZ* Kaya KILIÇTURGAY[5]

Depts. of infectious Diseases, and [2]Microbiology, Medical School of İnönü University, Malatya
[3]Dept. of Microbiology Medical School of Harran University, Şanlıurfa,
[4]Dept. of Infectious Disease Medical School of Atatürk University, Erzurum
[5]Dept. of Infectious Disease Medical School of Uludağ University, Bursa, TURKEY

Recombinant Hepatitis B vaccines have met some problems such as nonresponders. We assessed the efficacy of tetanus toxoid (TT) and Pre S2 + S containing recombinant hepatitis B vaccine (S2S RHB) in 10 nonresponders to S2SRHB. We applied simultaneous administration of S2SRHB and TT to ten persons (group I) who did not to respond (anti-HBs < 10 IU/L) after 3 doses of S2SRHB (given as 20 pgr : one dose, intramuscularly (IM), in months: 0-1-2). 6 nonresponders (group II) received 3 additional doses by the same route as the initial vaccination (S2SRHB, IM, 1 mo. Interval). In group I, after first S2SRHB +TTdose 2/10 (20%) responded with anti-HBs levels > or= 10 IU/L and after the second, another 3/10 (30%) responded and after the third, no more 0/10 (0 %) responded, corresponding to a total response rate of 50 %. In group II, after the first additional S2SRHB dose, 0/6 (0%) responded, with anti-HBs levels < 10 IU/L, after the second, 1/ 6 (16.66%) responded with anti-HBs level 15 IU/L and after third dose no additional nonresponse was noted. Both seroconversion rate and the antibody titer level for anti-HBs antibody were significantly higher in group I than in group II (p< 0.05). These results suggest that simultaneous administration of S2SRHB and TT is more effective in nonresponders than additional doses given by the same route as the initial vaccination. But further studies should be necessary including large, different nonresponder groups to confirm this conclusion. [Turk J Med Res 1997; 15(2):68-71]

Key Words: Hepatitis B vaccine, Tetanus toxoid, Nonresponder

Conventional hepatitis B vaccines, mainly consisting of the S gene product of the viral genome, have met several problems: They have induced vaccine escaped hepatitis B (HBV) mutants, antibody response after the vaccination is not prompt, and a fraction of vaccines do not produce antibodies at all (nonresponders) (1-3).

Several observations strongly argue that inclusion of the preS2 region may augment the immunogenisity and effectiveness of HBV vaccines (4-6).

Nevertheless some healthy personel or haemodialysis patients or hepatitis B carrier mothers' neonates are still nonresponded to Pre S2 + S containing recombinant hepatitis B vaccine (S2SRHB) (7-9). This may represent a mechanism of supression of neutralizing anti-hepatitis B virus antibody response, a phenomenon that accompanies the development of the chronic HBV-carrier state (10). The mechanisms causing nonresponsiveness to hepatitis B vaccines in humans remain largely unknown (11). Desombere et al (11) had shown that polymorph nuclear blood cells (PBMC) from nonresponders did not proliferate to HBsAg in-vitro, whereas they vigorously proliferated upon stimulation with tetanus toxoid.

The aim of this trial was to compare the antibody responses to S2SRHB vaccine and S2SRHB + TT in the nonresponders.

Received: Jan. 20, 1997 **Accepted:** April 29, 1997

Correspondence: Emine SÖNMEZ
Zafer mah. 14. Sok.
Sancak -1 Apt D: 16
Malatya, TURKEY

METHODS

A total 16 subjects were enrolled in the study, who ranged in age from 18 to 55 years (means 27.82 ± 6.14). They were negative for all HBV markers (ELISA Organon) and all had previously received at least three shots of S2SRHB (Genhevac, Pasteur) vaccines but nevertheless remained negative for anti-HBS antibody.

First nonresponder group (group I) contained 5 healthy and 5 hemodialysis patients. This group received S2SRHB vaccine (20 pgr: one dose.IM , in left deltoid muscle) and tetanus toxoid (TT) (1 ml_: 80 IU, IM, in right arm) (Adsorbed tetanus vaccine- Te Anatoxel Berna), simultaneously, at a 1 month interval.for consequent three months. Post vaccination blood samples

were obtained from all subjects at 1,2, and 4 months following the first vaccine.

Second nonresponder group (group II) contained 5 healthy persons and one haemodialysis patient. This group received 3 doses by the same route as the initial vaccination (S2SRHB, IM, 1 mo. Interval). Post vaccination blood samples were obtained from all subjects at 1,2, and 4 months following the first additional dose of vaccine.

All blood samples were studied for HBsAg, AntiHBc, Anti-HBs using ELISA (Organon). Serum transaminases, rhumatoid factors were tested in each serum sample to asses the biological safety of these vaccines.

For statistical analysis, x^2 test was used.

RESULTS

All participants' characteristics were shown on Table 1.

Anti-HBs titers exceeding 10 IU/mL generally were accepted as a protective titer. Anti-HBs responses to vaccination were shown on Table 2.

Anti-HBs titers at 4 months were shown on Table 3.

DISCUSSION

In previous studies, S2SRHB vaccine was highly immunogenic in adults . It induced antibodies to HBsAg at a protective level in more than 90 % of the subjects (1,12). Some persons still remained nonresponder to S2SRHB vaccine (7-12).

Several studies have been carried out and reports on the efficiency of these vaccines have been published.But it is obvious that these vaccines need improvement; especially in high-risk groups such as haemodialysis patients or heart transplant patients or healthy nonresponders (13,14).

There are some studies about simultaneous administration of diphteria / tetanus / pertussis / polio vaccines and hepatitis B vaccine in a simplified immunization programme (15,16). But there is no study about efficiency of TT to immune response of S2SRHB in nonresponders.

We found total response rate of 50 % to simultaneous administration of S2SRHB and TT in the nonresponders. And we found total response rate of 16.66 % to S2SRHB vaccine with additional three doses. There was statistically significance between two groups ($p< 0.05$).

Table 1. Characteristics of partipiciants

Character	Group I		Group II	
n	10	%	6	%
Age	18 to 55		18 to 55	
Age(mean ± SE)	26.82 ±5.14		28.80 ± 7.10	
Male/Female	6/4		3/3	
Healthy person	5	50	**5**	83.33
Haemodialysis patients	5	50	1	16.66
Previous vaccination (S2SRHB)	10	100	6	100
HBV markers (-)	10	100	6	100
AST/ ALT level (normal)	10	100	6	100
Rheumatoid factor	10	100	6	100

Table 2. Anti-HBs responses in the groups

	Anti-HBS IU/L											
	1 month				2 month				4 mouth			
Group	<10	%	>10	%	<10	%	>10	%	<10	%	>10	%
Group I*	8	80	2	20	5	50	5	50	5	50	5	50
Group II*	6	100			5	83.3	1	16.6	5	83.3	1	16.6

*p<0.05 Between group I and II for anti-HBs responses

Table 3. Anti-HBS titers at 4months in groups

Group	<10	%	10-50	%	50 to up	%
Group I*	5	50	4	40	1	10
Group II*	5	83.33	1	16.66	-	-

* p< 0.05, between group I and II for Anti-HBs titers

Struve J et al (9) found seroconversion after additional recombinant HB vaccine doses as 61% in the responded. Hemmerling et all (13) found approximately 76.2 % response with anti-HBs levels (> 10 IU/mL) for S2SRHB vaccine in nonresponder groups. These are higher than our results. This may depend on different group characteristics, different vaccine dose and route.

Haemodialysis patients had been vaccinated a random according to different vaccination schedules as 5 pgr, 10 pgr, 20 pgr, 40 pgr; and variations in the number of injections as well as the dose and type of vaccine did not result in clinically important enhancement of the anti-HBs response (17,18). We used same dose (20 pgr) vaccine in groups, because our groups were small and they received one dose hepatitis B vaccine.

These results indicate that simultanous S2SRHB + TT vaccination is remarkably effective in inducing antibodies in nonresponders and this route is more effective than additional S2SRHB vaccine. But further studies is necessary in large and different nonresponder groups.

Pre S2 ve S içeren rekombinant Hepatit B aşısının cevapsız grupta tetanoz toksoidi ile birlikte uygulandığı klinik bir çalışma: Bir ön çalışma

Rekombinant Hepatit B aşılarının aşıya cevapsızlık gibi bazı problemleri vardır. Biz Pre S2+S içeren recombinant hepatit B aşısına (S2SRHB) cevap vermeyen 10 nonresponder kişide S2SRHB ve Tetanoz toksoidinin (TT) birlikte uygulanmasının etkilerini değerlendirdik. Üç doz S2SRHB aşısına rağmen (20 pg: bir doz, İM, 0,1,2. Aylarda) antikor cevabı olmayan (anti-HBs <10 IU/mL) 10 kişide (grup I) S2SRHB ve TT'ni eşzamanlı uyguladık. Altı cevapsız kişide de (grup II) başlangıçtaki aşılama gibi 3 ilave doz (S2SRHB, bir ay ara ile) uygulandı. Grup I'de ilk S2SRHB+TT dozundan sonra 2/10 (%20)'u anti-HBs seviyesi 10 IU/mL'ye eşit veya üzerinde olacak şekilde cevap verdi; ikinci uygulamadan sonra diğer 3/10 (% 30) 'u verdi; üçüncü uygulamadan sonra cevap yoktu, toplam cevap oranı % 50 oldu. Grup H'de ilk ilave S2SRHB dozundan sonra cevap yoktu, ikinci ilave dozdan sonra 1/6 (%16.66)'sında 15 IU/mL anti-HBs seviyesinde cevap vardı, üçüncü ilave dozdan sonra cevap yoktu (0/6). Serokonversiyon ve antikor türeleri grup I'de Grup II'ye göre anlamlı şekilde yüksekti (p<0.05). Bu sonuçlar, aşıya cevapsız kişilerde, S2SRHB+TT'nin eşzamanlı uygulamasının ilave üç doz uygulamasına göre daha etkili olduğunu gösterir. Fakat daha büyük ve farklı gruplarda yapılan çalışmalarla bu sonucun doğrulanması gerekir.
TT Klin Araştırma 1997; 15(2):68-71]

REFERENCES

1. Akahane Y. Clinical trial of a preS2 - containing recombinant hepatitis B vaccine . In: Nishioka K, Suzuki H, Mishiora S, Oda T, eds. Viral Hepatitis and Liver Diseases. Tokyo, Berlin, Heidelberg, New York, London, Paris, Hong Kong, Barcelona, Budapest: Springer- Verlag , 1994; 533-5.

2. Carman WF, Zanetti AR, Karayiannis P, et al. Vaccine-induced escape mutant of hepatitis B virus . Lancet 1990; 336: 325-9.

3. Okamoto H, Yano K, Nozaki Y, et al. Mutations within the S gene of hepatitis B virus transmitted from mothers to babies immunized with hepatitis B immune globulin and vaccine. Pediatr Res 1992; 32: 264-8.

4. Neurath AR, Kent SBH, Parker K, et al. Antibodies to a synthetic peptide from the Pre S 120-145 region of the hepatitis B virus envelope are virus-neutralizing. Vaccine 1986; 4: 35-7.

5. Itoh Y, Takai E, Ohnuma H, et al. A synthetic peptide vaccine involving the product of the pre-S2 region of hepatitis B virus DNA; protective efficacy in chimpanzeas. Proc Natl Acad Sei USA 1986; 83: 9174-8.

6. Milich DR, Thornton GB, Neurath AR, et al. Enhanced immunogenicity of the pre-S region of hepatitis B surface antigen. Science 1985; 228: 1195-9.

7. George J, John GT, Jacop CK, et al. Active immunization against hepatitis B infection of a haemodialysis population. Nat Med J India 1994; 713: 115-6.

8. Canho R, Grosheide PM, Schalm SW, et al. Failure of neonatal hepatitis B vaccination : the role of HBV-DNA levels in hepatitis B carrier mothers and HLA antigens in neonates. J Hepatol 1994; 20 (4): 483-6.

9. Struve J, Aransson B, Frenning B, et al. Seroconversion after additional vaccine doses to nonresponders to three doses of intradermally or intramuscularly administered recombinant hepatitis B vaccine. Scand J Infect Dis 1994; 26(4); 468-70.

10-Bamaba V, Franco A, Albert! A, et al. Selective killing of hepatitis B envelope antigen-specific B cells by class I-restricted, exogenous antigen-specific T lymphocytes. Nature 1990; 345 (6272); 258-60.

H.Desombere I, Hauser P, Rossau R, et al. Nonresponders to hepatitis B vaccine can present envelope particles to T lymphocytes. J Immunol 1995; 154: 520-4.

12. Tron F, Degos F, Brechot C, et al. Randomised dose range study of a recombinant hepatitis B vaccine produced in mammalian cells and containing the S and pre-S2 sequences. J Infect Diseas 1989; 160 (2): 199-204.

13. Hemmerling AE, Müller R, Firusian N, et al. Clinical experience with pre-S2 containing Hepatitis B vaccine (HG-3) in different nonresponder groups. In: Nishioka K, Suzuki H, Mishiora S, Oda T (eds). Viral Hepatitis and Liver Diseases. Tokyo, Berlin, Heidelberg, New York, London, Paris, Hong Kong, Barcelona, Budapest: Springer- Verlag, 1994; 540-2.

14. Jungers P, Chauveau P, Laubaris T, et al. Immune response to hepatitis B vaccine in chronic uremic patients. In: Coursaget P, tong MJ, eds. Progress in Hepatitis B immunization (WHO colloque INSERM, vol 94, Labbey Eurotext Paris, 1990; 187-95.

15. Coursaget P, Yvonnet B, Relyveld EH, et al. Simultaneous administration of diphteria / tetanus / pertusis / polio vaccine and hepatitis B vaccine in a simplified immunization programme. Dev Biol Stand 1986; 65: 169-75.

16. Barone P, Mauro L, Leonardi S, et al. Simultaneous administration of HB recombinant vaccine with diphteria and tetanus toxoid and oral polio vaccine : a pilot study. Acta Pediatr Jpn 1991; 33(4) 455-8.

17. Simith-Leijs MBL, Kramer P, Heijtink RA, et al. Hepatitis B vaccination of haemodialysis patients: randomized controlled trial comparing plasma-derived vaccine with and without pre-S2 antigen. Eur J Clinical Investigation 1990; 20:540-5.

18. Stevens CE, Alter HJ, Taylor PW, et al. Hepatitis B vaccine in patients receiving hemodialysis. N Eng J Med 1984; 311:496-501.