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IMMUNOGENICITY OF A HEPATITIS B SUBUNIT VACCINE IN HEMODIALYSIS AND IN RENAL TRANSPLANT RECIPIENTS

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A hepatitis B subunit vaccine was given to 59 medical staff members, 106 hemodialysis patients and 28 renal allograft recipients. The vaccine consisted of formalin-inactivated hepatitis $B_{surface}$ antigen (HBsAg) and was given in 3 doses (times 0, 1 and 6 months) of 20–40 μg. Some of the vaccinees received anti-HBs antibodies together with the first vaccine dose (active/passive vaccination). One month after the last injection, 93% of the medical staff members who had received active/passive immunisation and 97% of those who had received active immunisation had detectable anti-HB_s antibodies with mean titers ranging from 1:512 to 1:1024. In the group of hemodialysis patients antibodies were detectable in 63–65% of the individuals who had received active or passive/active immunisation in mean titers between 1:32 and 1:64. Finally, only 32% of the renal allograft patients developed measurable anti-HBs antibodies, the titers of responders being still lower than in the hemodialysis patients. Side effects occurred following 10% of all vaccine injections and were always mild in nature.

Within the 12 months observation period following the first vaccination, 3 HBV events occurred in the 193 individuals: One aclinical case detected by a transient seroconversion against the hepatitis B core antigen, one anicteric and one icteric hepatitis case.

The data illustrate the difficulties for active immunisation against hepatitis B of hemodialysis patients or of renal transplant recipients.

hepatitis B vaccine; hemodialysis; renal transplant

INTRODUCTION

Various studies have shown that formalin-inactivated 22 nm hepatitis $B_{surface}$ antigen (HBsAg) subunit vaccines are highly immunogenic. 90–98% of healthy individuals develop antibodies against HBsAg (anti-HBs) after 3 injections [6,10,14,16,22]. In contrast, only 60–90% of vaccinated hemodialysis patients develop anti-HBs [2,15,21]. A deficient immune reactivity often associated with severe renal insufficiency might be the cause [4,13] of this decreased responsiveness.

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Vaccination also provides a high degree of protection against hepatitis B virus (HBV) infection. Protective efficacy has been demonstrated in a large double-blind trial with 1083 homosexuals [17], in smaller trials in medical personnel of hemodialysis centers [1], in hemodialysis patients [11], and in Senegalese children [12]. It was observed that HBV infections, if they occur at all in vaccinated individuals, do so either within the first weeks after initiation of vaccination or later or mainly in individuals who develop no or only low titered anti-HBs [18]. This observation further strengthens the assumption that vaccine induced anti-HBs protects against HBV infections.

Individuals with various types of immunodeficiency are especially prone to HBV infections. They not only acquire such infections more often than do healthy individuals, they also more frequently become chronically infected, and thus represent a source for further spread of HBV [5,19]. Hence, efficient vaccination of such patients would be highly desirable.

We report here on a comparative immunogenicity trial with a hepatitis B subunit vaccine in various high risk populations, including patients with renal allografts. The purpose of the study was to have good base line data for the development of new vaccination schedules for patients, whose immunological responses are likely to be suboptimal.

MATERIAL AND METHODS

Two lots of a highly purified formalin-inactivated vaccine (with alum adjuvant), consisting of the spherical 22 nm particles of HBsAg, developed by M.R. Hilleman and coworkers, and produced by Merck Sharp & Dohme were used (MSD lot numbers 65249/80700/C-6607 and 802/C-F733). The production procedure and the results of phase I and phase II trials of this vaccine are described in detail elsewhere [7,8]. Vaccination was offered to staff members, patients on hemodialysis, and patients with renal allografts of the renal units of the University Hospital Zurich, City Hospital Waid, Zurich, the Neumünster Hospital, Zollikerberg, and the Cantonal Hospitals of Aarau, Chur and Schaffhausen. Vaccine was given to the first 25–35 individuals in each group, who gave their written consent and who had no signs of past or ongoing infections with HBV. Amongst patients with renal allografts only those were accepted who were under steady-state medication with prednisone (10–20 mg), and azathioprine (50 mg) – a treatment status generally achieved 3–4 months after transplantation – and who had no graft rejection episode within the preceding 4–6 months. Amongst patients with end stage renal disease a distinction was made between those who were in a steady-state of hemodialysis and those who were newly admitted to hemodialysis. Also included were staff members and patients under hemodialysis of one center (City Hospital Waid) who had received hepatitis B immunoglobulin (HBs Ig, KABI) every 3–4 months in the preceding 8 months to 2 years [23]. Such individuals as well as newly admitted hemodialysis patients received a 'last' injection of HBs immunoglobulin (4 ml KABI or 3 ml MSD in newly admitted patients) together with the first vaccination (passive/active immunisation). Another group of hemodialysis patients, whose responses are reviewed here, received

vaccine but no HBs immunoglobulin (active immunisation). This group is part of a large European multicenter immunogenicity trial coordinated by Professor Deinhardt, Max von Pettenkofer Institute, Munich, F.R.G. Data were analysed for individuals who had been vaccinated three times, and who had reached month 9 (3 months after the third injection) by Nov. 1, 1981. The age and sex distributions of these groups and the modes of immunisation are given in Table 1. All individuals were vaccinated at times 0, 1 and 6 months. All patients received 40 μg of HBsAg protein per injection. The staff members were divided into two groups, one receiving 20 μg, and the other 40 μg doses per injection.

The following parameters were tested in every blood sample taken at intervals of 1–3 months: HBsAg (AUSRIA II, Abbott), anti-HBs (AUSAB, Abbott), anti-HBc (CORAB, Abbott), HBe and anti-HBeAg (radioimmunoassays, Abbott), and the transaminases SGOT and SGPT. Sera positive for anti-HBs were titrated in 2-fold dilution steps. The titers are expressed as the last serum dilution yielding a positive test (using program B of an ANSR-B gamma counter, Abbott).

Vaccine recipients recorded their own body temperature and any side effects for a period of 6 days following each injection. Furthermore, all doctors caring for such patients were urged to record any unusual events.

RESULTS

By Nov. 1, 1981, 9 or more months had elapsed since the initial vaccination in 193 individuals. 5 individuals had died. None of the deaths were in any way related to vaccination (e.g., 1 suicide, 1 brain tumor, 1 patient with decompensation of severe pre-existing cardiac insufficiency and final septicemia). 4 individuals were lost to follow-up.

Figures 1 and 2 summarize the occurrence and titers of anti-HBs at the various bleeding times. Of the staff members, 93–96% had anti-HBs after 7 months, while this was the case in only 57–65% of hemodialysis patients, and in 32% of patients with renal allografts. The average titers of those individuals who developed anti-HBs were between 1:512 to 1:1064 for staff members, between 1:32 and 1:64 for hemodialysis patients, and between 1:16 and 1:32 for patients with renal allografts. Of the staff members 73–85% had anti-HBs titers equal to or above 1:32 (130 IU). This was the case in 30–34% of patients on hemodialysis, but only in 18% of patients with renal transplants. Within vaccinee groups there were no significant differences in anti-HBs frequencies and titers between individuals receiving active vaccination and those given active/passive immunisation. There were also no obvious differences between males and females or between various age groups. Staff members responded as well to 20 μg doses of vaccine as to 40 μg.

Two patients with renal allografts were vaccinated despite pre-existing anti-HBc (without anti-HBe and anti-HBs). Both developed anti-HBs 30 days after the first vaccine dose, and titers were high when compared to other reactive patients of this collective.

TABLE 1

Frequency of positive anti-HBs responses, of side effects of HBV events during a 12-month follow-up period in 193 individuals vaccinated with HB subunit vaccine

Vaccinees						Percent with detectable anti-HBs				No. of individuals with at least one side effect	% Injections with side effect(s)	HBV events	Pathological liver enzymes without HBV events
Group	Mode of immunisation	Number ♂	♀	Total	Average age (years) (range)	month 3	month 6	month 7	month 12				
Patients with renal allografts	active	13	15	28	45 (22–64)	18	18	32	25	4	5	none	1[d]
Hemodialysis patients 'steady state'	active	19	41	60	53 (23–63)	28	42	63	63	12	8	1[a] (serologic only)	1[d]
	active/passive	12	13	25	51 (19–70)	(92)	57	65	52	4	9	1[b] anict. hepatitis 1[b] icteric hepatitis	5[e]
newly admitted	active/passive	10	11	21	45 (19–70)	–	36	57	55	2	7	1[c] icteric hepatitis	1[d]
Staff members	active	19	12	31	35 (22–59)	90	94	97	96	6	11	none	1[d]
	active/passive	9	19	28	38 (23–54)	(95)	86	93	93	9	25	none	1[d]
Total		82	101	193						37	10	1[a]+2[b]+1[c]	5[d]+5[e]

[a] Borderline positive (undiluted serum) anti-HBc level in one serum sample.

[b] Conversion to HBsAg positive at months 5 and 9; both patients without pre-existing HBsAg.

[c] HBeAg positive at time 0, HBsAg positive 14 days after first injection.

[d] In one serum sample of each patient: one of the two liver enzymes above normal (less than twice upper limit of normal).

[e] 2 patients with icteric hepatitis (nonA/nonB) (liver enzymes above 1000 U/l).
3 patients with anicteric hepatitis (nonA/nonB)

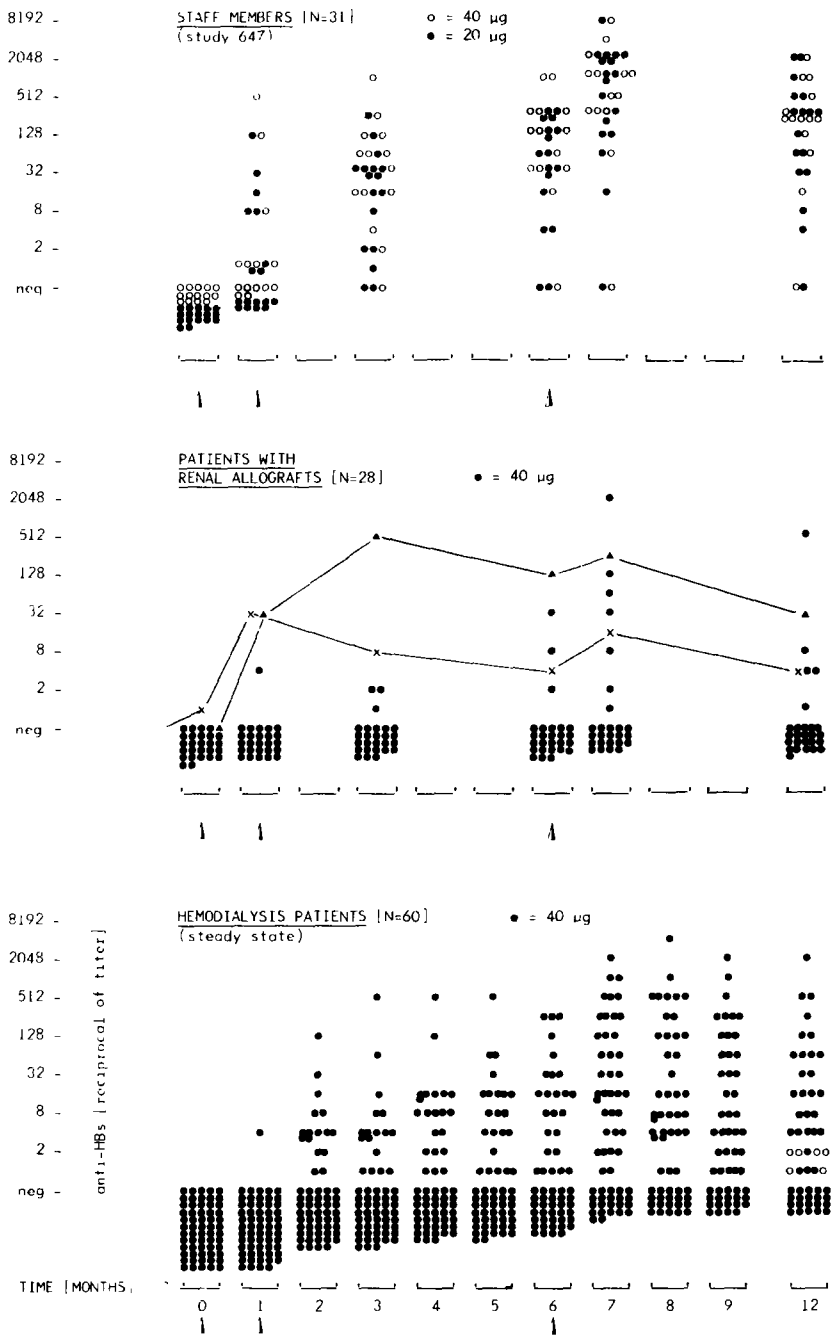

Fig. 1. Anti-HBs after active immunisation with hepatitis B vaccine. ▲ = vaccine injections; ○ ● = µg HBsAg per injection; ▲/X = individuals with preexisting anti-HBc alone. The titers given correspond approx. to international units: 1 : 4 = 17 IU; 1 : 32 = 130 IU; 1 : 256 = 1150 IU; 1 : 2048 = 8600 I.U.

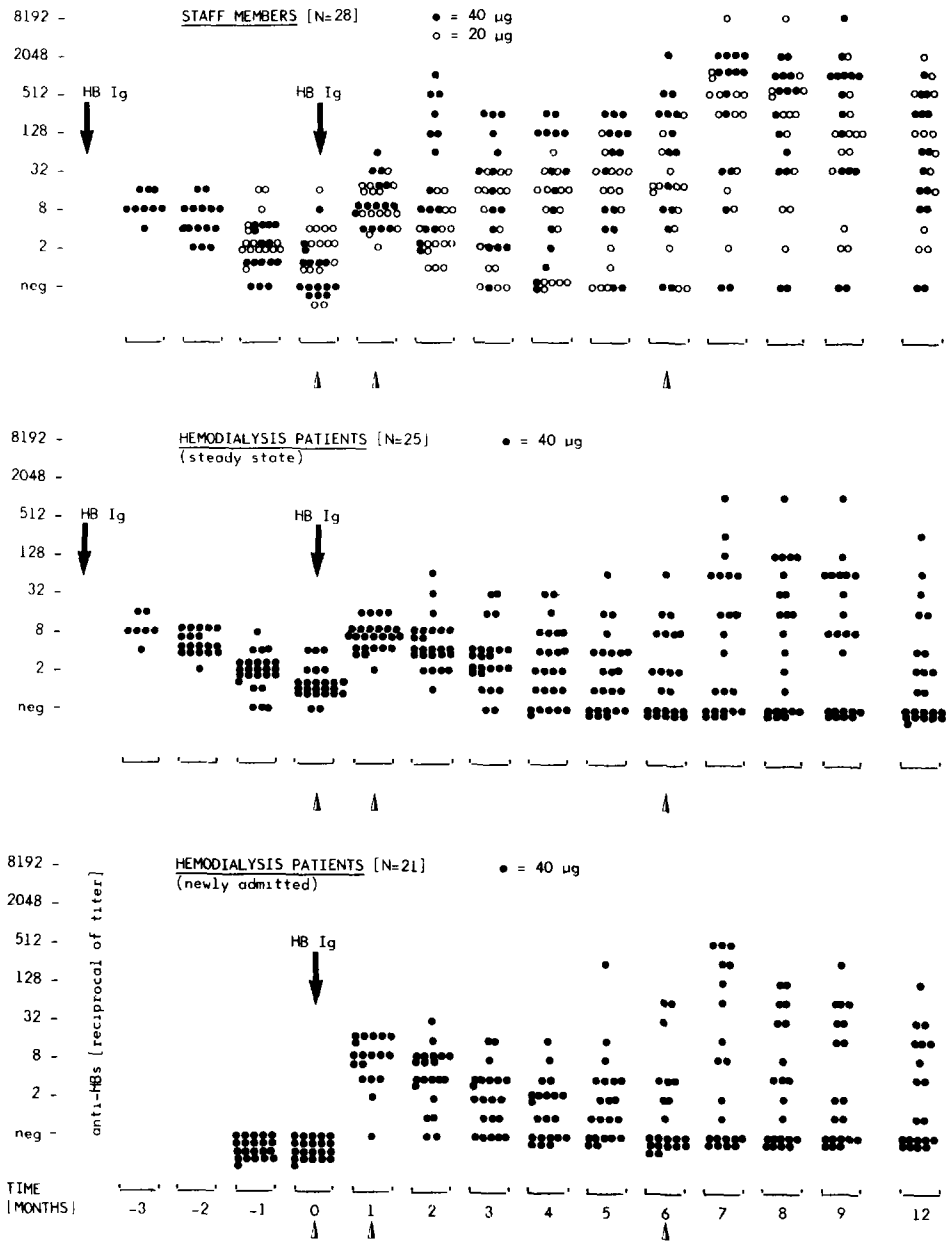

Fig. 2. Anti-HBs after active/passive immunisation with hepatitis B vaccine. ↓ = HB Ig: 3 ml of Hepatitis B immunoglobulin MSD or 4 ml HB Ig Kabi; ▲ = vaccine injections; ○ ●, µg HBsAg per injection.

Table 1 also summarizes the frequency of side effects, HBV events, and elevation of liver enzymes without specific signs of HBV infections: 20% (37 of 193) of all individuals vaccinated had at least one symptom which they themselves attributed to one of the three injections of the vaccine. 10% of over 650 injections of vaccine were thought by the vaccinated individuals to be followed by side effects. In only one instance did the side effects lead to withdrawal from the program. They consisted in order of frequency of: pain at the injection site, headache, nausea, malaise, neuralgic pain (usually but not exclusively in the proximity of the injection site), arthralgia/myalgia, vertigo, sweating, and strong thirst. Three individuals had episodes of fever within the 7-day period following one of the three injections, the temperature never exceeding 38°C.

After the initiation of vaccination, there were 4 HBV events amongst the 193 vaccinated individuals. In one hemodialysis patient anti-HBc was detected in one of the consecutive bleedings (month 4). He had no anti-HBs at that time, but such antibodies became detectable at month 7. Liver enzymes remained normal over the whole surveillance period. In another hemodialysis patient, anti-HBc was detected at month 5. A blood sample taken 10 days later showed HBsAg. This was still detectable at month 11. The patient was never jaundiced, but has continued to have slight elevation of liver enzymes. Another hemodialysis patient became jaundiced at month 9 and simultaneously became HBsAg positive. One newly admitted hemodialysis patient had HBeAg at time 0, HBsAg becoming detectable 10 days later. In this case, HBV infection must have occurred well before vaccination was initiated.

In 5 patients, one of the two liver enzymes showed slightly elevated values, but only in one of several consecutive bleedings. In no instance were clinical symptoms noted. High liver enzyme levels were observed within a two week period among 5 patients in one hemodialysis unit. Enzymes remained elevated for several weeks and in two patients exceeded 1000 IU. These 2 individuals became jaundiced. None of these 5 patients showed signs of a newly acquired infection with hepatitis A virus. Since other, non-vaccinated individuals within the same hemodialysis unit simultaneously developed anicteric hepatitis, it was inferred that a cluster of nonA/nonB hepatitis infections had occurred.

DISCUSSION

Our findings confirm and extend those reported by others [1,2,10]: (1) The MSD HB subunit vaccine is highly immunogenic. Approximately 95% of healthy individuals develop anti-HBsAg after 3 injections. (2) Hemodialysis patients develop anti-HBs less often and at lower titers than healthy individuals. (3) Simultaneous injections of HBs Ig and vaccine do not markedly hamper the formation of antibodies nor do they result in lower antibody titers [3,20]. (4) Side effects occur infrequently and are mild in nature.

A new finding is that only about one third of patients with renal allografts, who are under immunosuppressive medication, develop anti-HBs after 3 injections, and that these individuals have rather low titers. Similar results might be expected in other patients who

are under cytostatic regimens. The booster-like reaction occurring after vaccination in two patients with renal allografts, who had signs of past HBV infection (anti-HBc only), is a further confirmation of the rule that immunosuppressive regimens very strongly inhibit primary immune reactions, but affect secondary immune reactions to a lesser degree.

No analysis was made concerning the protective efficacy of the vaccine. The epidemiological background of the renal transplant and hemodialysis units participating in the study was that 20–30% of the hemodialysis patients and patients with renal allografts are positive for HBsAg [9].

An important problem to be solved comes with the non-reactors making up 3–7% of healthy individuals, 30–40% of hemodialysis patients, up to two thirds of patients with renal allografts and most probably also of all other patients under immunosuppressive treatment. New vaccination schedules are now being tested in new series of patients. In the present test series non-reactors have already been revaccinated or will be so in the near future. Preliminary results suggest that such additional injections of vaccine might not solve the problem. Thus, one conclusion can be made already now: patients whose diagnosis or disease course allows to predict a high risk of HBV infections, e.g. those who are likely to be subjected to immunosuppressive treatment or repeated surgery, should be vaccinated *before* such treatments are initiated. This holds especially true in areas where the attack rates for HBV infections are still considerably high.

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REFERENCES

1 Crosnier, J., Jungers, P., Couecoucé, A.M., Laplanche, A., Benhamou, E., Degos, F., Lacour, B., Prunet, P., Cerisier, Y. and Guesry, P. (1981) Randomised placebo-controlled trial of Hepatitis B surface antigen vaccine in French haemodialysis units: I. Medical staff. Lancet, 1, 455–459.

2 Crosnier, J., Jungers, P., Couecoucé, A.M., Laplanche, A., Benhamou, E., Degos, F., Lacour, B., Prunet, P., Cerisier, Y. and Guesry, P. (1981) Randomised placebo-controlled trial of Hepatitis B surface antigen vaccine in French haemodialysis units: II. Haemodialysis patients. Lancet, 1, 797–800.

3 Deinhardt, F., Zachoval, R., Schmidt, M., Frösner, H. and Frösner, G. (1981) Active and active-passive immunisation against Hepatitis B virus infection. in: Hepatitis B vaccine. (INSERM Symposium no. 18). Ed.: Maupas, P. and Guesry, P. Elsevier/North Holland Biomedical Press B.V., Amsterdam, pp. 167–171.

4 Goldblum, S.E. and Reed, W.P. (1980) Host defenses and immunologic alterations associated with chronic hemodialysis. Ann. Intern. Med., 93, 597–613.

5 Grob, P.J. (1980) The significance of Hepatitis B infection in patients on hemodialysis or with renal transplants. in: Proceedings of the European Symposium on Hepatitis B. Ed.: Krugman S. and Sherlock, S. Merck Sharp & Dohme International Division of Merck & Co., Inc., Rahway, N.J., pp. 39–53.

6 Hilleman, M.R., Bertland, A.U. and Buynak, E.B. et al. (1978) Clinical and laboratory studies of HBsAg vaccine. in: Viral Hepatitis. Ed.: Vyas, G.N., Cohen, S.N. and Schmid, R. Franklin Institute Press, Philadelphia, pp. 525–538.

7 Hilleman, M.R., Buynak, E.B. and Roehm, R.R. et al. (1975) Purified and inactivated human hepatitis B vaccine: Progress report. Am. J. med. Sci., 270, 401–404.

8 Hilleman, M.R., Buynak, E.B., McAleer, W.J., McLean, A.A., Provost, P.J. and Tytell, A.A. (1979) Newer developments with human hepatitis vaccines. J. Med. Virol., 4, 327–340.

9 Jost, R., Russi, E., Grob, P.J. and Binswanger, U. (1979) Hepatitisvirus-B-Infektion und Hepatopathie nach Nierentransplantation. Schweiz. Med. Wschr., 109, 1748–1756.

10 Maupas, P., Goudeau, A. and Coursaget, P. (1978) Immunization against Hepatitis B in man: a pilot study of two years' duration. in: Viral Hepatitis. Ed.: Vyas, G.N., Cohen, S.N. and Schmid, R., Franklin Institute Press, Philadelphia, pp. 539–556.

11 Maupas, P., Goudeau, A., Coursager, P., Drucker, J., Bagros, P., Baudin, S. and Geslin, N. (1978) Vaccine against Hepatitis B – 18 months prevention in a high risk setting. Med. Microbiol. Immunol., 166, 109–118.

12 Maupas, P., Chiron, J.P., Barin, F., Coursaget, P., Goudeau, A., Perrin, J., Denis, F. and Diop Mar, I. (1981) Efficacy of Hepatitis B vaccine in prevention of early HBsAg carrier state in children. Controlled trial in an endemic area (Senegal). Lancet, 1, 289–292.

13 Revillard, J.P. (1979) Immunologic alterations in chronic renal insufficiency. Adv. Nephrol., 8, 365–382.

14 Reesink, H.W., Reerink-Brongers, E.E., Brummelhuis, H.G.J., Lafeber-Schut, L.J.Th., van Elven, E.H., Duimel, S.J., Balner, H., Stitz, L.W., van den Ende, M.C., Feltkamp-Vroom, Th.M. and Cohen, H.H. (1981) Heat-inactivated HBsAg as a vaccine against Hepatitis B. Antiviral Res., 1, 13–25.

15 Seef, L.B., Wright, E.C. and Zimmerman, H.J., et al. (1978) Type B hepatitis after needlestick exposure: prevention with Hepatitis B immune globulin. Final report of the Veterans Administration Cooperative Study. Ann. Intern. Med., 88, 285–293.

16 Szmuness, W., Stevens, C.E., Harley, E.J., Zang, E.A., Taylor, P.E., Alter, H.J. and The Dialysis Vaccine Trial Group (1981) The immune response of healthy adults to a reduced dose of the Hepatitis B vaccine. J. Med. Virol., 4, 327–340.

17 Szmuness, W., Stevens, C.E., Harley, E.J., Zang, E.A., Oleszko, W.R., William, D.C., Sadovsky, R., Morrison, J.M. and Kellner, A. (1980) Hepatitis B vaccine. Demonstration of efficacy in a controlled clinical trial in a high-risk population in the United States. New Engl. J. Med., 303, 833–841.

18 Szmuness, W., Stevens, C.E., Zang, E.A., Harley, E.J. and Kellner, A. (1981) A controlled clinical trial of the efficacy of the Hepatitis B vaccine (Heptavax B); A final report. Hepatology (Baltimore), 1, 377–385.

19 Szmuness, W. (1979) Large-scale efficacy trials of Hepatitis B vaccines in the U.S.A.: Baseline data and protocols. J. Med. Virol., 4, 327–340.

20 Szmuness, W., Stevens, C.E., Oleszko, W.R. and Goodman, A. (1981) Passive-active immunization against Hepatitis B: Immunogenicity studies. Lancet, 1, 575–577.

21 Thomssen, R., Stamm, Br. and Gerlich, W. (1979) Simultaneous active-passive immunization of guinea pigs with HBIg and a HBsAg-vaccine. Preliminary Program, International Symposium on Viral Hepatitis, Munich, Federal Republic of Germany, April 5–7, 1979, p. 38.

22 Zachoval, R., Frösner, G. and Deinhardt, F. (1980) Impfungen gegen Hepatitis B. Ergebnisse einer Immunogenitätsstudie. Münch. Med. Wschr., 123, 1506–1508.

23 Zaruba, K. and Joller, H. (1981) Prevention of Hepatitis B in hemodialysis patients and staff using Hepatitis B immunoglobulin. Dialys. Transpl., 10, 210–212.