CLINICAL RESEARCH

Efficacy of a heat inactivated hepatitis B vaccine in male homosexuals: outcome of a placebo controlled double blind trial

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Abstract

The efficacy of a heat inactivated hepatitis B virus vaccine, containing 3 μg hepatitis B surface antigen (HBsAg), was studied in a high risk group of 800 susceptible homosexual men by a randomised placebo controlled double blind trial. At the trial end point (21·5 months), 17 hepatitis B virus infections had occurred in vaccinated subjects (attack rate 4·8%) and 56 in subjects receiving a placebo (attack rate 23·8%). This reduction in the incidence of hepatitis B virus infections in vaccinated subjects was highly significant ($p < 0·0001$). Two months after the first injection 72·3% of the vaccinated subjects had formed antibodies against hepatitis B surface antigen, and this percentage increased to 89% at four months. Maximum anti-HBs titres were reached five months after the first vaccination, the geometric mean titre being 107·6 mIU. Even vaccinated subjects with a low antibody response (≥ 1 and < 10 mIU) were found to be protected from HBsAg-positive infections. The vaccine had no serious side effects.

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Introduction

Male homosexuals are at high risk of acquiring a sexually transmitted hepatitis B virus infection.[1 2] Among a group of 2946 homosexuals living in the Netherlands—a country with a low prevalence of hepatitis B virus infection—we found serological evidence of a past or present infection in 60·3% and among 316 susceptible men an annual attack rate of 27·6%.[3] When a vaccine against hepatitis B virus infection, developed at the Central Laboratory of the Netherlands Red Cross Blood Transfusion Service,[4 5] became available for clinical trials we decided to study the efficacy of this vaccine among male homosexuals. The vaccine, which is heat inactivated and contains 3 μg of hepatitis B surface antigen (HBsAg) per dose, had been found to be safe and immunogenic in experimental animals and in a small group of human volunteers at low risk.[6 7] We report the outcome of the placebo controlled double blind study among homosexual men.

Subjects and methods

SELECTION OF PARTICIPANTS

Screening of sera from male homosexuals for hepatitis B virus markers was started in June 1980. Men without markers were invited to participate in the study and were given information about it during an interview. Details of their medical history and life style were recorded. Subjects were entered into the trial if they were between 16 and 50 years of age, were negative for HBsAg, antibodies to hepatitis B surface antigen (anti-HBs), and antibodies to hepatitis B core antigen (anti-HBc), had serum alanine transferase activity < 50 IU/l and no serious illness, and had had at least two different male sexual partners in the preceding six months. Participants had to give their informed consent.

VACCINE AND PLACEBO

The vaccine was prepared from a pool of plasma containing HBsAg, subtypes ad and ay.[5] Inactivation was achieved by heating for 90 seconds at 101°-104·5°C. The heat inactivated material was absorbed to aluminium phosphate and pasteurised for 10 hours

at 65°C. One dose of the vaccine contained 3 μg HBsAg adsorbed to 0·6 mg aluminium phosphate. Quality control was performed according to the requirements proposed by the World Health Organisation.[8] One dose of the placebo consisted of 600 μg human albumin adsorbed to 0·6 mg aluminium phosphate. Vaccine and placebo were indistinguishable and were stored at 4°C.

CONDUCT OF THE TRIAL

Subjects were randomly assigned to either the vaccine or placebo group. The trial was conducted double blind. Three injections of vaccine or placebo were given intramuscularly at monthly intervals. Subjects were asked to note side effects, which were recorded at their next visit. Blood was collected every month during the first five months and every three months thereafter. A person was considered lost to follow up when he failed to attend for longer than six months.

Each blood sample was tested for the presence of HBsAg and anti-HBc and for serum alanine transferase activity. In addition the first blood sample (month 0) was tested for anti-HBs and hepatitis A virus antibody (anti-HAV). Measurements of anti-HBs in the second and subsequent blood samples were not made available until the code of the trial was broken. If a blood sample was found to be positive for HBsAg or anti-HBc, or both, or had a serum alanine transferase activity $\geqslant 50$ IU/l (upper normal limit 20 IU/l) the subject was asked to return for confirmation of results and follow up.

An ethics committee evaluated the study protocol and decided on the date when the code should be broken. An independent doctor to whom hepatitis B virus infections were reported was the only one who had access to the code. Medical representatives of the Dutch organisation for homosexual men and women were informed about the study.

DEFINITION OF HEPATITIS B VIRUS INFECTIONS

The hepatitis B virus infections were classified by an independent committee before the code of the trial was broken and without access to the measurements of anti-HBs.

Definite infections—HBsAg or anti-HBc or both were present in at least three sequential blood samples. (1) Hepatitis B: HBsAg present in at least one blood sample and serum alanine transferase activity $\geqslant 21$ IU/l in at least two sequential blood samples taken one to three weeks apart with at least one sample with activity $\geqslant 50$ IU/l. (2) HBsAg-positive infection: HBsAg present in at least one blood sample. (3) Anti-HBc-positive infection: only HBc present, HBsAg always absent.

Possible infections—Anti-HBc was present in two sequential blood samples, followed by one or more samples in which HBsAg and anti-HBc were absent.

LABORATORY METHODS

Tests for the presence of HBsAg, anti-HBs, and anti-HBc were performed by radioimmunoassay (Abbott Laboratories). The anti-HBs titres were expressed in mIU according to the method described by Hollinger.[9] Serum alanine transferase activity was measured by an automated kinetic method. Total antibodies to hepatitis A virus were measured by an enzyme linked immunosorbent assay (Organon).

TABLE I—*Comparison of characteristics of subjects in vaccine and placebo groups*

Characteristics	Vaccine group	Placebo group
No entering trial	397	403
Mean age (years) (SE)	30·7 (0·35)	30·1 (0·35)
Mean duration of homosexuality in years (SE)	11·4 (0·35)	11·5 (0·35)
% With >10 different sexual partners in preceding 6 months	34·5	35·5
% With history of jaundice	14·5*	8·7*
% With history of syphilis	18·5	15·8
% With hepatitis A antibody	39·2	40·3
% Living in city with >500 000 inhabitants	64·0	69·5
Interval between first and second injections (days) (SE)	30·5 (0·22)	30·7 (0·28)
Interval between first and third injections (days) (SE)	61·6 (0·71)	60·4 (0·34)
No failing to return for follow up (%)	19 (4·8)	16 (4·0)

*$p < 0.025$.

STATISTICAL METHODS

All data were stored in a computer. For calculation of the life table attack rate the actuarial method was used.[10] Survival rates were compared according to the method of Desu.[11] Other statistical methods used were the χ^2 test, Student's t test, analysis of variance, and the Z test using Greenwood's formula.

Protection by the vaccine was expressed as the protective efficacy rate (attack rate with placebo minus attack rate with vaccine divided by attack rate with placebo $\times 100$). The geometric mean titre was calculated for vaccinated subjects who developed anti-HBs $\geqslant 1$ mIU at the time stated.

Results

Between 10 November 1980 and 31 December 1981, 835 men entered the trial. Because of the presence of markers for hepatitis B at the time of the first injection, 35 men were excluded; of the remaining 800 subjects in the study, 397 received the vaccine and 403 the placebo. For most characteristics both groups were similar (table I); only the percentage of men giving a history of jaundice was higher in the vaccine group ($p < 0.025$).

The first, second, and third injections were given to 397, 390, and 384 recipients of the vaccine and to 403, 401, and 396 recipients of the placebo respectively. The interval between the first and second injections was 30·7 days (SE 0·28) in the placebo group and 30·5 (SE 0·22) in the vaccine group (target 30·5 days). The interval between the first and third injections was 60·4 (SE 0·34) and 61·6 (SE 0·71) days in the placebo and vaccine groups respectively, the target being 61 days. During the trial 19 subjects in the vaccine group and 16 in the placebo group were lost to follow up, mostly owing to lack of motivation or because they moved.

INFECTION RATE IN BOTH GROUPS

When the code of the trial was broken on 1 September 1982, 69 definite and four possible hepatitis B virus infections had occurred among the subjects (fig 1). Among the placebo group 56 subjects had hepatitis B virus infections, most (31) having HBsAg-positive infections. In the vaccine group 17 subjects had hepatitis B virus infections, of whom nine were positive for HBsAg.

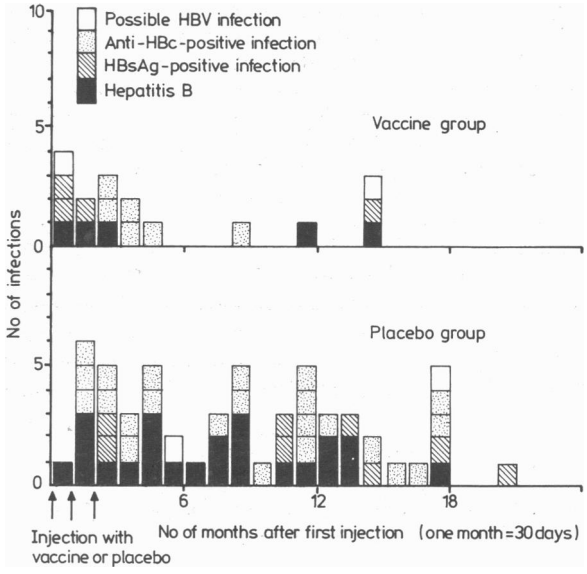

FIG 1—Hepatitis B virus infections among subjects in vaccine and placebo groups.

For all the different categories of hepatitis B virus infection an appreciable reduction in the attack rate among the vaccine group was evident at the end of the trial (table II). For all HBsAg-positive infections with HBsAg the attack rate at the end point (645 days) was 2·7% in the vaccine group compared with 16·9% in the placebo

TABLE II—*Comparison of life table attack rates at trial end point (over 645 days) of different categories of hepatitis B virus infections in vaccine and placebo groups with the protective efficacy rate (comparison from the time of the first injection)*

Category of infection	Placebo group		Vaccine group		Probability	Protective efficacy rate (%)
	No of subjects	Attack rate (%) (±SE)	No of subjects	Attack rate (%) (±SE)		
Hepatitis B*	23	7·7 (±2·0)	5	1·5 (±0·7)	<0·001	80·7
All HBsAg-positive infections	31	16·9 (±6·7)	9	2·7 (±0·9)	<0·003	84·3
Anti-HBc-positive infections	23	7·4 (±1·6)	6	1·6 (±0·6)	<0·005	78·5
All definite infections	54	23·1 (±6·4)	15	4·2 (±1·1)	<0·0001	81·8
All infections (possible infections included)	56	23·8 (±6·3)	17	4·8 (±1·2)	<0·0001	79·7

*Serum alanine transferase activity ⩾50 IU/l.

group ($p < 0·003$), for anti-HBc-positive infections 1·6% compared with 7·4% ($p < 0·005$), and for all hepatitis B virus infections 4·8% compared with 23·8% ($p < 0·0001$). For the different categories the protective efficacy rate varied between 78·5% and 84·3%, being 79·7% for all hepatitis B virus infections together at the trial end point. The curve of the attack rate among the vaccine group levelled off rapidly when compared with the attack rate among the placebo group (fig 2).

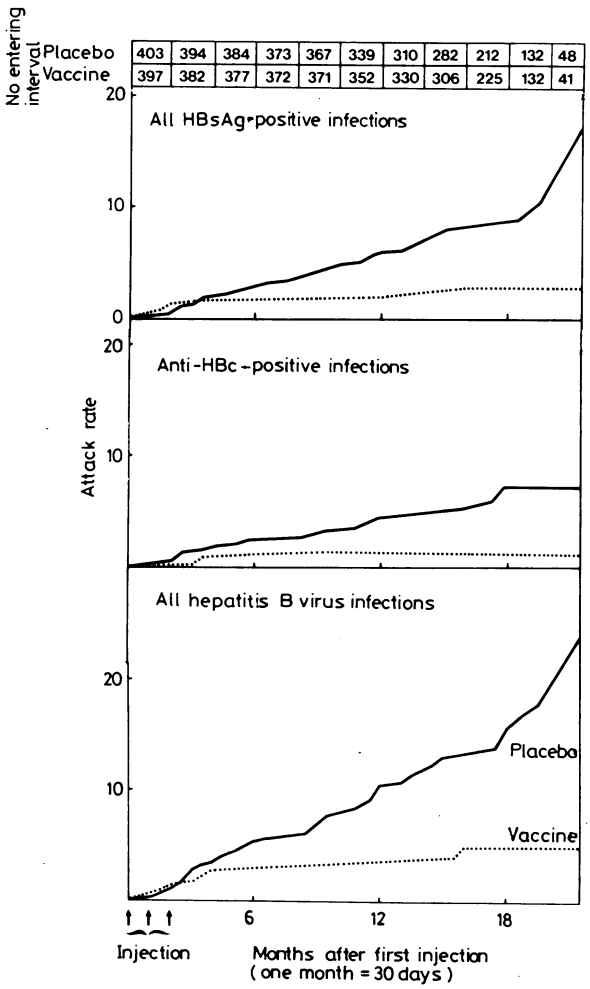

FIG 2—Comparison of life table attack rates of hepatitis B virus infections in subjects in vaccine (. . .) and placebo (——) groups.

ANTI-HBS RESPONSE IN VACCINE GROUP

One month after the first vaccination 33·1% of subjects had developed anti-HBs, usually at low titres (geometric mean titre 3·5 mIU, SE 1·11) (fig 3). After the second and third injections the percentage of vaccinated subjects who developed anti-HBs increased to 72·3% at month 2 and 87·4% at month 3. At month 4 a maximum of 89% had demonstrable anti-HBs (46·3%, ⩾100 mIU; 31%,

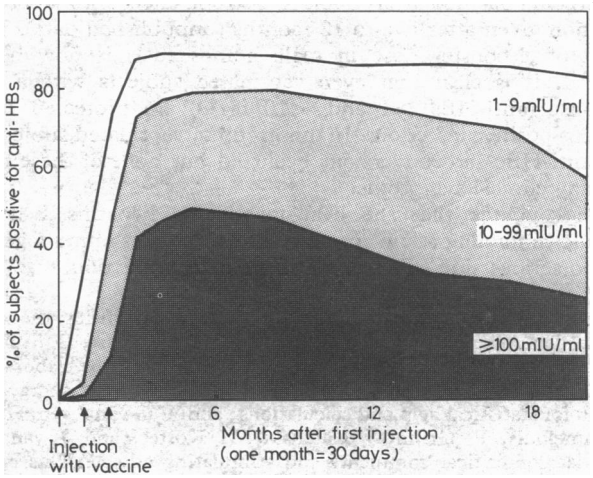

FIG 3—Anti-HBs response of subjects in vaccine group in mIU/ml.

between 10 and 100 mIU; and 11·7%, ⩾1 and <10 mIU) whereas 11% had not formed antibodies. There were a few low responders (⩾1 and <10 mIU) who lost anti-HBs during follow up, so that at month 17, 85·7% still had anti-HBs (30·5%, >100 mIU; 39·5%, between 10 and 100 mIU; and 15·7%, ⩾1 and <10 mIU). The geometric mean titre (mIU) of vaccinated subjects positive for anti-HBs increased from 17·1 mIU (SE 1·10) at month 2 to a maximum of 107·6 mIU (SE 1·10) at month 5 and gradually declined thereafter to 47·1 mIU (SE 1·12) at month 17 ($t = 35·45$; $p < 0·005$).

Relation to hepatitis B virus infections

Among the vaccine group nine HBsAg-positive, seven anti-HBc-positive, and one possible hepatitis B virus infection occurred. All nine HBsAg-positive infections were found in subjects who had not (yet) formed anti-HBs. The seven anti-HBc-positive infections and the possible hepatitis B virus infection occurred in vaccinated subjects who had low titres of anti-HBs (⩾1 and <10 mIU) after one or more vaccinations.

SIDE EFFECTS OF VACCINE

The vaccine had no serious side effects. After the first injection 65·4% of vaccinated subjects recorded soreness at the injection site compared with 44·5% among the placebo group (χ^2 test, $p < 0·001$); after the second and third injections these figures were respectively 39·8% compared with 24·7% ($p < 0·001$) and 32·1% compared with 15·1% ($p < 0·001$). The soreness subsided within one to two days.

Discussion

This randomised placebo controlled double blind study shows that the heat inactivated hepatitis B vaccine, containing a low dose of HBsAg (3 μg), gives a high degree of protection for all categories of hepatitis B virus infections. Two other studies of the efficacy of a vaccine among susceptible male homosexuals have recently been published. Szmuness *et al*, using the 40 μg formaldehyde inactivated vaccine (Merck), found a protective efficacy rate at the trial end point (735 days) of 81·4% for all hepatitis B virus infections including anti-HBc seroconversions.[12 13] In the Centers for Disease Control multicentre trial with 20 μg Merck vaccine a protective efficacy rate (end point 510 days) of 55·9% was found,[14] whereas in our study the rate (end point 645 days) for all hepatitis B virus infections together was 79·7%.

In our study 89% of the vaccinated subjects formed demonstrable anti-HBs. In the other two studies using the Merck vaccine a higher percentage of responders was found with a 40 μg dose (96%) but a slightly lower one with a 20 μg dose (85%). Studies with the heat inactivated vaccine among patients undergoing haemodialysis point to a strong effect with a booster injection given after five to 12 months (unpublished data). The effect of a booster dose in male homosexuals is now being studied. It is clear that even vaccinated subjects with a low response (anti-HBs $\geqslant 1$ and <10 mIU) are protected from HBsAg-positive infections. In this group of vaccinated subjects a few anti-HBc seroconversions occurred but none of these men had any clinical symptoms.

We conclude that the heat inactivated hepatitis B virus vaccine containing a low dose of HBsAg (3 μg) is immunogenic, has no serious side effects, and offers good protection.

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ONE HUNDRED YEARS AGO Most of the French hospitals, according to our contemporary *Engineering*, have now a photographic studio attached, for photographing the sick persons at different times. The rapid dry plate process is employed, and Professor Charcot, of the Salpêtrière, has devised an electrically worked camera, which is very useful in taking a series of views in rapid succession. Certain classes of patients are photographed on their entry into the hospital, and at regular intervals afterwards. In cases of hysteria, for example, it is interesting to note the original contractions, and compare them with succeeding ones. The photographs are placed in an album for study of the disease, and comparison with others taken from other patients. In this way the leading features of the disease will be recognised. The new printing processes also enable these photographs to be copied, and distributed to other hospitals and medical men. Micro-photography, or the photographing of microscopic objects, is also a valuable branch of hospital-work which is becoming better recognised every day. The apparatus of Professor Charcot consists of a camera with a movable aluminium shutter, controlled by an electro-magnet and clockwork. A key and battery send an electric current through the magnet at the will of the operator, and working the shutter exposes an objective to the object. The photographer controls the apparatus by his hand, and, with his finger on the key, watches his patient until the desired moment arrives, then presses his finger, and exposes the plate for the instant required to take the likeness. When a regular series of views in rapid succession are required, the hand-key is replaced by an insulating barrel, set round with metal contact-pieces like a commutator, and these pieces make contact with a contact-spring as the barrel revolves by clockwork. These metallic pieces are made of a triangular form, so that, when a short exposure is required, the spring is placed so as to rub over them towards the apices of the pieces; and, when a longer exposure is required, it is caused to rub over them near the bases. The metronome, or automatic mercury current-interruptor, of M. Gaiffe is also applicable to this camera as an automatic key. (*British Medical Journal*, 1883;ii:1084.)

ONE HUNDRED YEARS AGO It is somewhat surprising that attention has not before been called in the House of Commons to the sewer-emanations which must make themselves unpleasantly perceptible to the olfactory nerves of members on their way to the House. There is one sewer-grating in the middle of the road immediately opposite the gates of St. Stephen's that must offend even the most hardened nose; and yet it was not until Thursday last week that Mr. J. G. Talbot felt emboldened to inquire of the President of the Local Government Board, whether his official virtue was not shocked by the existence of such a state of things. Sir Charles Dilke's reply was studiously obscure. He was careful not to explain, as he might have done, that the sewers in the neighbourhood of Whitehall are laid at such a level that there is practically no chance of any reasonably rapid flow of sewage along them. This has, to some extent, been the cause of the unsanitary state of the new Government offices, to which attention was recently again directed at Liverpool. It does not clearly appear from Sir Charles Dilke's reply who are the "authorities" responsible for looking after these sewers; but it is certainly a remarkable commentary on our London system of Government, that such a state of affairs should exist in the place where beyond all others sanitary perfection might have been expected. (*British Medical Journal*, 1883;ii:384.)