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Meningococcal Infections *

3. Studies of Group A Polysaccharide Vaccines

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Studies with 4 lots of group A meningococcal polysaccharides in 458 adult human volunteers showed that such vaccines are nontoxic and highly immunogenic. Comparisons of jet injection and subcutaneous (needle) methods of administration showed that both were equally immunogenic.

The safety and efficacy of group C meningococcal polysaccharide vaccines for the prevention of group C disease have been demonstrated by several large-scale field studies in US Army recruits (Artenstein et al., 1970a; Gold & Artenstein, 1971). Experience with group A meningococcal polysaccharide vaccines in man has been much less extensive. Gotschlich, Goldschneider & Artenstein (1969a) presented data on 5 volunteers who received 50 μg of group A polysaccharide. A prompt, group-specific antibody response was observed in each subject. Subsequently, 53 recruit volunteers were immunized without ill effect; all but 2 of 51 tested showed an antibody response (Gotschlich, Goldschneider & Artenstein, 1969b).

The present studies provide information on the safety and immunogenicity of 4 lots of purified group A polysaccharide vaccines in more than 450 additional human volunteers.

MATERIALS AND METHODS

Subjects

In accordance with US Army regulations, informed consent was obtained from all volunteers. The majority of subjects were active-duty US Army personnel; a few were laboratory workers.

Vaccine

Group A polysaccharides were prepared by the method of Gotschlich, Liu & Artenstein (1969). Lot A-5, produced at the Walter Reed Army Institute of Research, was previously described by Gotschlich, Goldschneider & Artenstein, (1969a, 1969b). Lot A-6 was prepared by the Department of Biologics Research, Walter Reed Army Institute of Research; Lots A-7 and A-8 were made by the Squibb Institute for Medical Research under a contract with the US Army Medical Research and Development Command.

Serology

Serum specimens were collected aseptically and stored at −20°C. Haemagglutinating antibody was assayed by methods previously described by Gotschlich, Liu & Artenstein, (1969) in which fresh human erythrocytes were sensitized with purified group A polysaccharide. The fluorescent antibody test employed was that described by Goldschneider, Gotschlich & Artenstein (1969) in which whole organisms served as the antigen. Fluorescein-conjugated rabbit antiserum against human heavy-chain IgG [1] was used.

RESULTS

Toxicity studies of lots A-6 and A-7 in man

A group of 5 laboratory volunteers was inoculated intracutaneously with 50 μg of lot A-6 in a volume of 0.2 ml. Erythema at the injection site began at 3–5 hours following the injection, measured 10–20 mm in diameter at 24 hours, and had disappeared within 48 hours. No tenderness was noted.

* From the Department of Bacterial Diseases, Walter Reed Army Institute of Research, Washington, D.C., USA.

[1] Supplied by Hyland Laboratories, Los Angeles, Calif., USA, and by Behring Diagnostics, Woodbury, N.Y., USA.

This lot was subsequently tested in 42 volunteer recruits who received the vaccine subcutaneously in a 50-μg dose. Only 2 subjects complained of gastrointestinal distress and diarrhoea beginning 6 hours after vaccination and were asymtomatic after 24 hours. No local reactions were seen.

A total of 21 subjects submitted urine specimens prior to, and at 24 and 48 hours after, vaccination. The only abnormalities noted were traces of albumin in 1 specimen from each of 3 subjects at random intervals. In all, 22 subjects tested before and after immunization showed no significant changes in haematocrit reading or in white cell or red cell morphology. Platelet estimations in peripheral smears from 6 subjects were normal.

Lot A-6 was tested for pyrogenicity in another group of 44 army volunteers. Vaccine was given subcutaneously as a 50-μg dose. Temperatures were recorded with an oral thermometer prior to, and at 18, 25, and 35 hours following, vaccination. There were no local or systemic reactions to the vaccine and no significant changes in any subject's temperature.

Vaccine lot A-7 was initially tested in 7 laboratory volunteers and administered subcutaneously in doses of 50 μg. All subjects were examined periodically for 48 hours and no local or systemic reactions were observed.

The complete blood count of each volunteer was recorded prior to, and at 24 and 48 hours following, vaccination. No consistent or significant changes were noted in any of the parameters tested.

Serological response to group A vaccines

Haemagglutinating antibody response was tested in 42 subjects who received lot A-6; 35 (84%) showed 4-fold or greater titre increases. All 7 volunteers who received lot A-7 vaccine showed 4-fold or greater haemagglutinating antibody response.

Dose titration of lot A-7

A total of 101 recruit volunteers in a basic-training company were divided into 3 groups, each of which received a different dose of vaccine—namely, 10, 50, or 100 μg—administered subcutaneously. There were no adverse reactions to these injections.

Serological responses measured by haemagglutination are shown in Table 1. Antibody responses to 50 μg and 10 μg doses were much greater than those to doses of 10 μg at each test period. Expressed in another way (Table 2) only 1 of 68 volunteers failed to show a 4-fold or greater rise in titre following the 50 μg or 100 μg dose whereas 5/33 (15%) failed to respond to the 10 μg injection. From this table it is also apparent that a significant proportion of subjects who received the lowest dose failed to show an antibody rise within 2 weeks but developed increasing titres over the next 6 weeks.

Table 1. Antibody response to lot A-7 vaccine

Dose group (μg)	Geometric mean haemagglutination titre ($\log_2$) at the following weeks:			
	0	2	4	8
10	0.45	2.9	3.5	3.6
50	0.32	5.6	5.9	5.6
100	0.29	5.3	5.3	5.7

Table 2. Number of subjects who failed to respond to lot A-7 vaccine

Dose group (μg)	No. failures[a]/No. tested at the following weeks after vaccination:			
	2	4	8	Total
10	14/33	9/31	5/31	5/33
50	2/36	0/35	0/35	0/37
100	3/30	1/30	1/27	1/31

[a] Less than 4-fold antibody titre rise.

Carrier surveys on this company showed that over 40% of the subjects were nasopharyngeal meningococcal carriers by the 4th week of the study but no group A strains were identified. Group A haemagglutinating antibody response was uninfluenced by carrier status.

Comparison of immunogenicity of three lots of group A polysaccharide vaccines

A total of 188 recruit volunteers in a basic-training company were divided into several groups, each receiving a different lot of vaccine. For each group the dosage was 50 μg injected subcutaneously. Altogether, 46 subjects received vaccine that had been stored under special conditions and these data are not included. The remaining subjects received vaccine

lot A-5, A-6, or A-7: these lots were stored cold in the lyophilized state and were hydrated just before they were administered. In all, 99 of the subjects provided serum specimens, which were assayed at the same time. Table 3 shows the results of haemagglutinating antibody tests on serum obtained prior to vaccination and 2 and 7 weeks later. These data show that the three lots were equally immunogenic. Peak haemagglutination titres occurred at 2 weeks with no significant change over the next 5 weeks. Of 10 subjects who failed to show an increase in titre at 2 weeks, only 4 showed such an increase at 7 weeks; in each instance the rise was only 4-fold, the minimum change considered significant.

Table 3. Serological response to three lots of group A meningococcal polysaccharide vaccine

Vaccine lot No.	Mean haemagglutinating antibody rise (log_2) at the following weeks:		No. failures [a]/No. tested at the following weeks:	
	2	7	2	7
A-5	3.8	3.8	2/33 (6 %)	1/33 (3 %)
A-6	3.7	3.9	4/32 (12 %)	2/32 (6 %)
A-7	3.5	3.6	4/34 (12 %)	3/32 (9 %)

[a] Less than 4-fold titre rise.

Table 4 illustrates the relationship of the haemagglutination prevaccination titre to the subsequent response to polysaccharide injection. Altogether, 6 of the 10 men who showed no response or a delayed antibody rise had prevaccination haemagglutination titres of 1 : 16 or greater. Out of 89 subjects who showed a positive response at 2 weeks, 87 had prevaccination titres of 1 : 8 or less.

A number of sera that showed no antibody rise in the haemagglutination test were retested by the fluorescent antibody test, which confirmed the lack of serological response.

Comparison of subcutaneous (needle) and jet injection routes of inoculation

Vaccine lot A-8 in 50-μg doses was injected into recruit volunteers by two different routes. One group of 36 subjects received the inoculum subcutaneously by needle and syringe in the deltoid region; another group of 35 subjects was immunized with automatic jet injection apparatus.[1] Serum specimens obtained prior to, and 1 and 2 weeks following, vaccination were tested for antibody rise by haemagglutination tests. The results are shown in Table 5. Geometric mean haemagglutinating antibody titres were essentially the same in both groups.

Table 5. Antibody response following subcutaneous and jet injection immunization with lot A-8 meningococcal polysaccharide vaccine

Group	Geometric mean haemagglutination titre (log_2) at the following weeks:			No. subjects with no rise in titre
	0	1	2	
Subcutaneous:				
titre	0.7	4.3	5.0	6 (17 %)
no. of subjects	36	17	36	
Jet injector:				
titre	1.5	4.1	4.7	6 (17 %)
no. of subjects	35	20	34	

[1] Supplied by the Scientific Equipment Manufacturing Corporation, Larchmont, N.Y., USA.

Table 4. Relation of prevaccination haemagglutinating antibody titre to vaccine response

Antibody response to vaccination	No. of subjects with the following prevaccination titres:							
	<1 : 2	1 : 2	1 : 4	1 : 8	1 : 16	1 : 32	1 : 64	1 : 128
positive: [a]								
2 weeks	30	26	23	8	2	0	0	0
7 weeks	1				2	1		
negative	2		1		1		1	1

[a] 4-fold or greater.

DISCUSSION

Purified meningococcal group A polysaccharide vaccines have been shown to be free of toxicity in 458 volunteers, and also previously in 58 subjects (Gotschlich, Goldschneider & Artenstein, 1969a, 1969b). Titration of one lot (A-7) of polysaccharide in volunteers showed that the antibody response to a 10-μg dose was inferior to those resulting from 50-μg, 100-μg injections in terms of magnitude of serum antibody and number of subjects who responded. However, 85% of subjects showed a significant haemagglutinating antibody rise even to the suboptimum 10-μg dose. Antibody response to 100 μg of vaccine was not significantly greater than that following a dose of 50 μg. Although peak titres were usually reached within 2 weeks following vaccination, about 6% of subjects who received 50 μg or 100 μg did not show a 4-fold titre increase until 4 weeks later. This delayed response might, in part, explain the 17% of serological failures observed in the jet injector experiment in which only prevaccination and 2-week post-vaccination sera were tested. Comparison of three different lots of group A vaccine given to volunteers as a 50-μg dose showed no significant differences in geometric mean haemagglutination titres or in percentage of serological conversions. Antibody response to the injection of group A polysaccharide was delayed or absent more often in subjects whose prevaccination haemagglutination titre was 1 : 16 or greater than in those with low baseline titres. Comparisons showed that both routes of inoculation, subcutaneously by needle and syringe or by jet injector apparatus, were equally immunogenic.

Experience with purified meningococcal group A polysaccharides has been similar to that previously reported for group C polysaccharides, 4 lots having been free of toxicity; the optimum dose appears to be 50 μg and all lots seem to be equally immunogenic in man (Artenstein et al., 1970b). Field studies are now indicated to determine the efficacy of the group A vaccines in the prevention of menigococcal disease.

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RÉSUMÉ

INFECTIONS MÉNINGOCOCCIQUES: 3. ÉTUDE DE VACCINS POLYSACCHARIDIQUES DU GROUPE A

On a expérimenté sur 458 volontaires quatre lots différents de vaccin antiméningococcique contenant des antigènes polysaccharidiques du groupe A (lots A-5, A-6, A-7 et A-8).

Aucun des vaccins n'a provoqué de réactions secondaires notables. Les lots A-5, A-6 et A-7 ont fait preuve d'un pouvoir immunogène équivalent: chez plus de 90% des sujets vaccinés, on a observé une hausse de 4 fois ou plus des titres d'anticorps hémagglutinants dans les 4 semaines suivant l'injection. Des doses de 50 et 100 μg du lot A-7 ont suscité une réponse significativement plus élevée qu'une dose de 10 μg; cependant, 85% des sujets vaccinés par cette dose faible ont élaboré des anticorps.

Les résultats de la vaccination n'ont pas varié selon que le vaccin était administré par la voie sous-cutanée classique ou à l'aide d'un injecteur sous pression.

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