

## A controlled field trial of a serogroup A meningococcal polysaccharide vaccine\*

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*A controlled field trial of a serogroup A meningococcal polysaccharide vaccine was conducted at three locations in Egypt during the winter cerebrospinal meningitis (CSM) season of 1971-72. The study population consisted of schoolchildren 6-15 years of age. No cases of serogroup A meningococcal CSM occurred in the group of students vaccinated with the test vaccine whereas 8 cases occurred in the control group vaccinated with tetanus toxoid, and 151 cases occurred in an unvaccinated contrast group. The case rate was significantly different between the test and control groups as well as between the test and contrast groups but was similar between the control and contrast groups. The previously demonstrated safety of the vaccine was confirmed. A significant serological response was elicited in the majority of the vaccinated students.*

The control of epidemic cerebrospinal meningitis (CSM) became possible after the introduction of sulfa drugs (Jusatz, 1959; Kent, 1969; Lapeyssonnie, 1968), but since 1963 this control has been endangered, especially in rural Africa, by the appearance and spread of sulfa-resistant meningococcal strains (Alexander et al., 1968; Millar et al., 1963; Vandekerkove et al., 1969). Active immunization has been suggested as an alternative control measure, but the available vaccines have generally proved ineffective (Cvjetanović, 1971). Recently, a poly-

saccharide extracted from serogroup C meningococci was shown to confer excellent protection against homologous serogroup C meningococcal disease in man (Artenstein et al., 1970; Devine et al., 1970). This polysaccharide was immunologically specific. A similar preparation was also prepared from serogroup A meningococci, the serogroup mainly responsible for African CSM as well as past epidemics in Europe and North America (Kent, 1969; Lapeyssonnie, 1963, 1968). This serogroup A polysaccharide vaccine was also shown to be both safe and antigenic in man (Gotschlich et al., 1969; 1972a; 1972b). A controlled field trial of its efficacy was therefore conducted at three locations in Egypt during the winter endemic CSM season of 1971-72.

\* This study was conducted under the auspices of the Ministry of Public Health of the Arab Republic of Egypt and of the World Health Organization. The opinions expressed herein are those of the authors and do not necessarily reflect the official views of the sponsoring authorities and organizations. The mention of proprietary products is for identification only and does not imply recommendation.

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### MATERIALS AND METHODS

#### *Plan of the trial*

The plan for the study was drawn up by a special committee at the Ministry of Health of Egypt. After the committee had examined the technical, ethical, and other aspects of the trial, it drew up a protocol, which was then discussed with health and school authorities in Cairo and Alexandria. When the plan had been accepted by the authorities, the public was fully informed before being asked to take part in the study. The same committee supervised the

execution of the study. Prior to the trial in Egypt, the vaccine had already been tested on schoolchildren and proved to be safe under those conditions.

The study was conducted as a controlled field trial following the general principles previously described (Cvjetanović, 1961; Pollock, 1966). The population was divided by systematic allocation of school classes into two study groups, a CSM vaccine group and a control group, who received, respectively, serogroup A meningococcal polysaccharide and tetanus toxoid. These vaccines were coded "blue" and "red", respectively, and their identity was unknown to the investigators, the field workers, and the vaccinated persons.

The efficacy of the trial vaccine was measured by assessing the incidence of CSM. A limited serological study was conducted concurrently, and carrier studies were also performed. Particular care was taken to ensure the precise diagnosis of CSM cases and to include enough subjects so that the number of cases would permit a valid statistical evaluation.

#### *Trial locality*

Three cities were selected for the trial—Alexandria, Cairo, and Giza—as they provided an adequate population of students. In this population CSM was known to be endemic, annually recurring, seasonal, and predictable. The selected cities had experienced several hundred CSM cases in each previous season. The commonest bacterial agent of CSM was *Neisseria meningitidis*, serogroup A.

#### *Trial population*

The study was conducted on schoolchildren 6–15 years of age. Approximately 75% of the students attending selected schools were included in the study making a total of 124 349 children vaccinated. They were divided into two groups, 62 295 receiving the serogroup A polysaccharide vaccine and 62 054 the control, the tetanus toxoid vaccine. The vaccinated children numbered 50 874 in Alexandria, 45 564 in Cairo, and 27 911 in Giza. The age and sex distribution of the two vaccinated groups was very similar, the mean ages varying by only 0.1 year (10.51 and 10.41) and median ages by merely 0.01 year (9.51 and 9.52). The schoolchildren were from similar socio-economic backgrounds, and division between the two groups was approximately equal within each school. Other factors such as food sources, living conditions, and medical facilities were similar for each group.

#### *Endemic meningococci*

Cultures obtained from carriers before and during the 1971–72 endemic season yielded meningococci from most of the recognized serogroups. While the majority of CSM was caused by meningococci of serogroup A, rough meningococci were the predominant strains in the carriers. In Cairo and Giza, serogroup A meningococci were predominant among infected individuals. In Alexandria, however, serogroup B meningococci were second in importance and serogroup A fell significantly to third position (Table 1). The case/carrier ratio was similar at the three locations, being 1/496 in Alexandria, 1/1770 in Cairo, and 1/1370 in Giza. Other meningococcus strains were isolated from CSM cases as follows: group B from 3 cases; C from 3; Y from 2; and W-135 from 1 case. No rough strains were isolated from CSM cases.

#### *Vaccine and vaccination procedure*

The meningococcal polysaccharide was prepared as described by Gotschlich et al. (1969, 1972b) from a culture of *N. meningitidis*, serogroup A, strain M-1027. A single dose consisted of 50 µg contained in 0.5 ml of diluent. The tetanus toxoid control dose was also 0.5 ml. Both vaccines were in a dry form.<sup>1</sup> They were transported and stored at or below –20°C before reconstitution and use. To test for degradation, vaccine samples obtained before shipment, upon arrival in Egypt, and following completion of the trial were compared for reduction of polysaccharide molecular weight (Gotschlich et al., 1972a). The initial molecular weight of approximately 170 000 remained essentially unchanged. Furthermore, inhibition of radioactive antigen binding was accomplished as readily with the vaccine before packaging as with all samples returned from the field. Therefore, it was concluded that no detectable degradation of vaccine occurred during the vaccination period.

Vaccinations were performed by teams of Egyptian physicians and technicians using hypodermic jet injectors.<sup>2</sup> The vaccines were reconstituted at the vaccination site as required, and only the amount of vaccine estimated to be needed for daily use was

<sup>1</sup> The meningococcus vaccine and the tetanus toxoid control were prepared and donated by the Mérieux Institute, Lyon, France.

<sup>2</sup> Ped-O-Jet (foot-operated) injectors with the standard nozzle for subcutaneous injection, F41-42; Scientific Equipment Manufacturing Corporation, Lodi, N.J., USA.

Table 1. Serogroup-specific carrier rates among students vaccinated with tetanus toxoid control vaccines

Rank <sup>a</sup>	Serogroup	Alexandria (306 subjects)		Serogroup	Cairo (296 subjects)		Serogroup	Giza (300 subjects)	
		No.	%		No.	%		No.	%
1	Rough	115	37.6	Rough	156	52.7	Rough	111	37.0
2	B	77	25.2	A	52	17.6	A	48	16.0
3	Y	23	7.5	B	30	10.1	Y	30	10.0
	A	20	6.5	Y	27	9.1	Multi-	25	8.3
	Multi- <sup>b</sup>	19	6.2						
	X	16	5.2						
4	Z	8	2.6	C	4	1.4	Z	13	4.3
	C	7	2.3	Z	3	1.0	B	12	4.0
				Multi-	3	1.0	X	9	3.0
				X	2	0.7	C	8	2.7
				D	2	0.7			
5	D	0	—				D	0	—
Totals	positive	285	93.1		277	93.6		256	85.3
	negative	21	6.9		19	6.4		44	14.7

<sup>a</sup> Ranking is based on 90 % confidence limits.

<sup>b</sup> " Multi- " = multiple agglutinating meningococcus strains.

withdrawn from storage on each occasion. The environmental temperature was between 15° and 20°C.

As far as possible, the two vaccines were given to alternate classes within each school grade. All the students in one class received one vaccine while all those in the next class received the other vaccine. Since students were not selectively assigned to classes, this procedure tended to ensure even vaccine distribution, in view of the large numbers involved. This system also provided advantages in vaccination efficiency and ensured that the correct vaccines were given. These factors facilitated double checking of the identity and vaccination status of subsequent CSM cases.

The vaccinations were administered in Alexandria from 14 to 27 December, in Giza from 22 December to 18 January, in Cairo from 3 to 18 January, and again in Alexandria from 6 to 9 February. Vaccinations were completed in the early phase of the seasonal increase in the number of CSM cases (Fig. 1).

#### Case finding and follow-up

It was essential to find and accurately identify all CSM cases in the two vaccinated groups. This was facilitated by an established procedure that every suspected CSM case should be immediately transferred to one of the government fever hospitals at Alexandria, Abbassia (Cairo), or Embaba (Giza). This occurred whether the patient was first seen by a private physician or at a public clinic. Furthermore, school health authorities were asked to report any child who was absent from the study schools more than 3 days in succession. At admission, the patient's identity and school affiliation were determined. These were compared with the data on a serially numbered card, colour-coded for vaccine type, that had been prepared for each vaccinated child at the time of vaccination. The identity of each patient was also checked by referring to school class records and was further confirmed with his family.

Bacteria were isolated from cerebrospinal fluid (CSF) and identified as described by Sanborn (1969). In addition, the immuno-osmophoresis (IOP) test

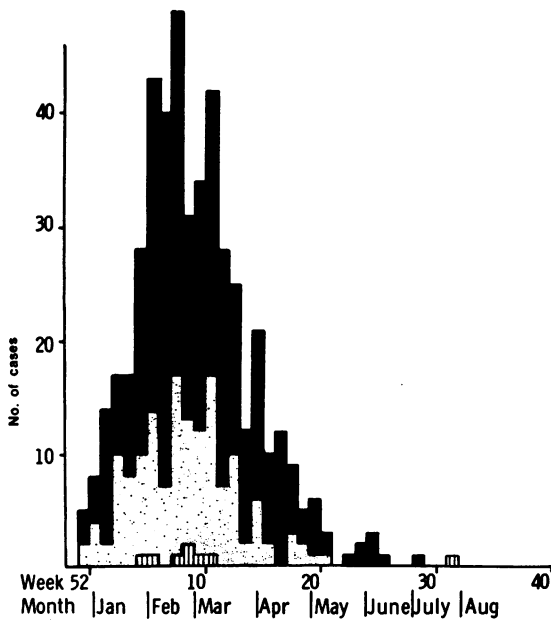


Fig. 1. The development of the epidemic of cerebrospinal meningitis caused by *Neisseria meningitidis* Group A in Alexandria, Cairo, and Giza during the 1971-72 season. Black = nonvaccinated adults; stippled = nonvaccinated students (contrast group); vertical lines = control students (tetanus toxoid).

was used to detect and identify meningococcal antigens in CSF (Edwards, 1971). Whenever possible, serum specimens were obtained at both the acute and convalescent stages for diagnostic confirmation.

#### Serological methods

Serum specimens were obtained from individuals of both vaccine groups before vaccination and 3 weeks after vaccination. These sera were analysed by the radioactive precipitin method (Gotschlich et al., 1972a). In addition, the IOP test was used to identify precipitating antibody in the sera of convalescent patients. For the purposes of statistical comparison, the observation period of the study was terminated at 31 July, about 6 months after the vaccinations were administered, the date falling well within the interepidemic period (Fig. 1).

#### Statistical methods

The chi-square test was used to compare group characteristics, the data being considered as a  $2 \times C$  contingency table. Differences in mean ages between treated and control groups by sex and locality were tested using the Z-test, where the means and standard

deviations were calculated from grouped data. Student's t-test was employed to examine the significance of the difference between mean antibody concentrations of paired sera and nonpaired mean antibody concentrations (Snedecor & Cochran, 1967). The significance of the difference between CSM case rates was tested by the zI-test (Langley, 1970).

## RESULTS

### Morbidity

Eight cases of serogroup A meningococcal meningitis occurred in the vaccine study population, all in the control group that received tetanus toxoid (Table 2). The resultant difference in case rates between subjects immunized with the meningococcal vaccine and those vaccinated with tetanus toxoid was statistically significant ( $P = 0.004$ ).

All patients displayed typical clinical signs and symptoms of CSM, and they all survived with no sequelae. In 4 cases the definitive diagnosis was made by isolation of serogroup A meningococci from CSF. In 2 others the CSF was culture-negative, but serogroup A meningococcal antigen was specifically detected and identified in the CSF by the IOP test. The diagnosis was confirmed serologically in the remaining 2 patients, whose sera contained specific serogroup A precipitating antibody at the convalescent stage; 1 serum was also tested by the HA test and was positive.

Four other CSM cases occurred in the study groups, 1 being in the serogroup A vaccine group (Table 2). This case was caused by *N. meningitidis*, serogroup B. The other cases were pneumococcal meningitis, tuberculous meningitis, and meningitis of unknown etiology, respectively.

Nonvaccinated students were designated as a "contrast group". There were 151 cases of serogroup A CSM among 1 403 508 subjects in this group, a case rate of 1.076 per 10 000. This rate was not significantly different from that of the control group ( $P = 0.62$ ). This in a way confirmed that the control group was representative of the non-immunized student population. When the case rate for the contrast group was compared with that for the serogroup A vaccine group, the difference was highly significant ( $P = 0.009$ ).

All the CSM cases in the control group occurred at least 17 days after vaccination. The age range of these patients was 9-15 years, with a mean of 10.9 years. These patients included 7 males and 1 female, the proportion of males being higher than that among the nonvaccinated students.

Table 2. Cerebrospinal meningitis cases among vaccinated students

City	Vaccination		Patient		No. of days between onset and vaccination	Causal organism	Diagnostic criteria
	Vaccine	Date	Age (years)	Sex			
Cairo	control	18 Jan	15	F	17	<i>N. meningitidis</i> Gr. A	immuno-osmophoresis
Giza	control	12 Jan	11	M	26	<i>N. meningitidis</i> Gr. A	serology (IOP) <sup>a</sup>
Alexandria	control	25 Dec	12	M	56	<i>N. meningitidis</i> Gr. A	culture isolate
Cairo	control	3 Jan	11	M	55	<i>N. meningitidis</i> Gr. A	immuno-osmophoresis
Giza	control	9 Jan	14	M	51	<i>N. meningitidis</i> Gr. A	culture isolate
Alexandria	control	7 Feb	9	M	28	<i>N. meningitidis</i> Gr. A	culture isolate
Cairo	control	16 Jan	10	M	59	<i>N. meningitidis</i> Gr. A	culture isolate
Alexandria	control	8 Feb	7	M	171	<i>N. meningitidis</i> Gr. A	serology (IOP; HA) <sup>a</sup>
Cairo	group A	4 Jan	9	F	18	<i>N. meningitidis</i> Gr. B	culture isolate
Cairo	control	5 Jan	6	F	21	<i>Diplococcus pneumoniae</i>	culture isolate
Giza	control	4 Jan	9	M	40	unknown	clinical findings
Cairo	control	9 Jan	9	M	23	<i>Mycobacterium tuberculosis</i>	clinical findings

<sup>a</sup> IOP = immuno-osmophoresis; HA = passive haemagglutination.

*Vaccine reactions*

The safety of this vaccine and the fact that it produced relatively few local reactions had previously been clearly demonstrated (Gotschlich et al., 1972a, 1972b; Sanborn et al., 1972). This trial confirmed those findings. In Alexandria, 78 vaccinated students were carefully observed for local vaccine reactions, the examinations being carried out on a double-blind basis. At 4 h after vaccination, there was erythema 1–2 cm in diameter and oedema at the vaccination site in 25 of the students given serogroup A polysaccharide vaccine and in 7 of those given tetanus toxoid vaccine. By 24 h there was still some local reaction in 4 of the former and in 1 of the latter. By 48 h, no local reactions could be found. There was pain in the arm immediately after vaccination, but this subsided quickly.

No generalized reactions occurred in either vaccine or control groups. One case of a suspected generalized reaction was reported, but investigation revealed that this child was a patient with rheumatic heart disease who had tonsillitis at the time and who had also received a typhoid-fever vaccination a few days previously. Therefore his illness was not attributed to the experimental vaccine; he recovered completely.

*Serological response*

Paired serum specimens from vaccinated students (86 in Alexandria, 51 in Cairo, and 6 in Giza) were

tested for precipitating antibodies against serogroup A meningococcal polysaccharide. From each location, 12 pairs were randomly chosen from those vaccinated with tetanus toxoid, and the remainder of the sera tested were from students vaccinated with serogroup A polysaccharide vaccine. The initial mean antibody levels in the Alexandria subjects were significantly lower than those of the Cairo or Giza students, which were generally the same. Mean antibody concentration was at least tripled following vaccination with serogroup A polysaccharide; this difference was highly significant ( $P < 0.001$ ). There was no significant increase in the mean antibody concentration in sera from students vaccinated with tetanus toxoid.

Group	Mean serum antibody level (µg/ml)		P
	prevaccination	postvaccination	
<i>Alexandria</i>			
vaccine	3.24	9.80	<0.001
control	1.68	1.62	>0.50
<i>Cairo</i>			
vaccine	5.34	21.95	<0.001
control	6.10	5.67	>0.50
<i>Giza</i>			
vaccine	7.28	24.45	<0.001
control	6.31	6.96	>0.50

The increase in mean antibody concentration was generally in proportion to the initial titre; however, some individuals with relatively low initial titres subsequently developed titres as high as or higher than the maximum actually measured (40 mg/ml). In Cairo and Giza, 28% and 22% of the vaccinated students, respectively, attained antibody levels of 40 mg/ml or higher, while in Alexandria, only 3% did.

#### DISCUSSION

The results of this field trial indicate that this polysaccharide vaccine prepared from serogroup A meningococci reduced to nondetectable limits the occurrence of CSM caused by serogroup A meningococci. Since the vaccine and control groups were basically comparable, the significant reduction in case rates appears to be attributable to the vaccine. Since 1 meningococcal CSM case of heterologous etiology occurred in a student in the serogroup A polysaccharide vaccine group, the vaccine may well be immunologically specific. Although the data indicated complete protection, the number of CSM cases was too small to determine the exact degree of protection. However, other studies with a serogroup C meningococcal vaccine prepared in a similar way indicated the protective effect of that vaccine to be approximately 90%. Therefore it may be assumed that the degree of protection offered by this serogroup A vaccine would be similar.

The majority of the students in the serogroup A vaccine group responded serologically, while no such serological responses were seen in subjects vaccinated with tetanus toxoid. The mean antibody increase was consistent with that found in studies of West African children (Gotschlich et al., 1972a).

The initial mean antibody levels in the vaccinated students in Alexandria were significantly lower than

those in the Cairo or Giza students. Since it has been shown that antibody production is stimulated by the carrier state (Sanborn & Vedros, 1966) this difference in titre probably represented the generally lower level of serogroup A carrier infection noted in Alexandria. The case rates of CSM caused by serogroup A meningococcus in the three areas were comparable, so the antibody level as measured by this test did not provide an indication of the levels of immunity in the communities.

The excellent safety record of this polysaccharide vaccine was confirmed. Local reactions were both transient and mild. Thus, the vaccine should be acceptable to populations at risk of contracting CSM.

The successful outcome of this field trial serves to indicate not only that the vaccine is effective, but that an immunization operation of this magnitude in the field is feasible. This success suggests that public health authorities elsewhere in Africa and in other places threatened by CSM epidemics should consider using serogroup A polysaccharide vaccine as a control measure. Further trials are in progress to confirm and extend these findings and to assess the duration of immunity and the modification of carrier rates.

Three main factors contributed to the successful mass vaccination programme: the use of the jet injector, proper storage and transport of the vaccine, and well organized national health services. Serogroup A polysaccharide vaccine requires subzero storage temperatures for maximum stability. Commercial freezers were used and proved to be satisfactory. Countries with health services comparable to those in Egypt, able to provide trained field vaccination teams and proper vaccine storage, should be capable of conducting a similar vaccination programme.

#### ACKNOWLEDGEMENTS

The authors are deeply grateful to the many health officials, laboratory technicians, and public health field workers whose untiring efforts made the study possible and to physicians in the Public School Administration and the Department of Epidemiology, High Institute of Public Health, for distributing the vaccine.

Operational and logistic support was provided by the High Institute of Public Health in Alexandria, the Central Public Health Laboratories in Cairo, US Naval Medical

Research Units No. 1 and 3 in Berkeley and Cairo, and the Environmental and Preventive Medicine Unit No. 7 in Naples.

This study was supported by grants from the World Health Organization, project M4305-01-1005BFG6 of the US Navy Bureau of Medicine and Surgery, and the Director's Fund of the US Naval Biomedical Research Laboratory. The vaccine was donated by the Institut Mérieux, Lyon, France.

## RÉSUMÉ

ESSAI PRATIQUE CONTRÔLÉ D'UN VACCIN POLYSACCHARIDIQUE  
CONTRE LES MÉNINGOCOQUES DU SÉROGROUPE A

En raison de l'incidence croissante de souches de *Neisseria meningitidis* résistantes aux sulfamides au cours des dernières années, les vaccins suscitent un regain d'intérêt pour la prévention de la méningite cérébro-spinale (MCS). Un vaccin polysaccharidique purifié s'est montré actif contre les infections à méningocoques du sérotype C et on a mis au point un vaccin similaire contre les méningocoques du sérotype A. Ce dernier vaccin a fait l'objet d'un essai contrôlé destiné à vérifier son efficacité.

L'essai a été conduit en 1971/72 en Egypte où la MCS due aux méningocoques du sérotype A est endémique et où on dispose de facilités pour les vaccinations de masse, la surveillance épidémiologique et les examens de laboratoire. Il a porté sur une population d'écoliers, âgés de 6 à 15 ans, des villes d'Alexandrie, du Caire et de Giza. Deux groupes ont été constitués: 62 295 enfants ont été vaccinés par le vaccin antiméningococcique et 62 054 (groupe témoin) ont reçu de l'anatoxine tétanique. En outre, 1 403 508 enfants (groupe de contraste) n'ont reçu aucune vaccination.

On n'a constaté aucun cas de MCS à méningocoques du sérotype A dans le groupe vacciné; 8 cas sont survenus dans le groupe témoin et 151 dans le groupe de contraste, soit une différence statistiquement significative entre le groupe vacciné et le groupe témoin ( $P = 0,004$ ) ainsi qu'entre le groupe vacciné et le groupe de contraste ( $P = 0,009$ ). L'innocuité du vaccin, déjà démontrée antérieurement, a été confirmée. Il n'y a eu aucune réaction secondaire généralisée, et les réactions locales ont été bénignes et passagères. On a enregistré une réponse immunitaire notable chez la plupart des vaccinés. Il existait une corrélation entre les titres pré-vaccinaux d'anticorps anti-méningocoques A et la fréquence des porteurs de germes.

Les moyens opérationnels mis en œuvre lors de cette étude ne diffèrent pas de ceux qu'il faudrait employer pour lutter contre la MCS dans d'autres pays d'Afrique. Les campagnes de vaccination de masse à l'aide de ce vaccin sont possibles et elles contribueraient efficacement à la prévention des épidémies de MCS due aux méningocoques du sérotype A.

## REFERENCES

- Alexander, C. E. et al. (1969) *Science*, **161**, 1019  
 Artenstein, M. S. et al. (1970) *New. Engl. J. Med.*, **282**, 417  
 Cvjetanović, B. (1961) *J. Hyg. Epidem. (Praha)*, **5**, 7  
 Cvjetanović, B. (1971) In: *Proc. Symposium on Bacterial Vaccines* Zagreb, Yugoslav Academy of Sciences & Arts, pp. 281-280  
 Devine, L. F. et al. (1970) *Amer. J. Epidem.*, **92**, 25  
 Edwards, E. A. (1971) *J. Immunol.*, **106**, 314  
 Gotschlich, E. C. et al. (1969) *J. exp. Med.*, **129**, 1349  
 Gotschlich, E. C. et al. (1972a) *J. clin. Invest.*, **51**, 89  
 Gotschlich, E. C. et al. (1972b) In: *Progress in immunobiological standardization*, vol. 5, Basle, Karger, p. 485  
*(Proceedings of the Twelfth International Congress for Microbiological Standardization, Annecy, 1971)*  
 Jusatz, H. J. (1959) *Cerebrospinal meningitis, 1927-1958*. In: *World atlas of infectious diseases, Part III*, 4th ed., Falk Verlag, Hamburg, p. 9  
 Kent, D. C. (1969) *J. Egypt. publ. Hlth Ass.*, **44**, 225  
 Langley, R. (1970) *Practical statistics*, revised ed., London, Pan Books, p. 245  
 Lapeyssonnie, L. (1963) *La méningite cérébro-spinale en Afrique*, Geneva, World Health Organization  
 Lapeyssonnie, L. (1968) *Med. trop.*, **28**, 709  
 Millar, J. W. et al. (1963) *J. Amer. med. Ass.*, **186**, 139  
 Pollock, T. M. (1966) *Trials of prophylactic agents for the control of communicable diseases*, Geneva, World Health Organization (*Monograph Series* No. 52).  
 Sanborn, W. R. (1969) *J. Egypt. publ. Hlth Ass.*, **44**, 385  
 Sanborn, W. R. et al. (1972) In: *Progress in immunobiological standardization*, vol. 5, Basle, Karger, p. 497  
*(Proceedings of the Twelfth International Congress for Microbiological Standardization, Annecy, 1971)*  
 Sanborn, W. R. & Vedros, N. A. (1966) *Health Lab. Sci.*, **3**, 111  
 Snedecor, G. W. & Cochran, W. G. (1967) *Statistical methods*, 6th ed., Ames, Iowa State University Press  
 Vandekerckove, M. et al. (1969) *Bull. Wld Hlth Org.*, **41**, 843

## ADDENDUM

Since completion of this report, 4 further cases of Group A meningococcal meningitis were observed in the study population up to the end of June 1973. All were in the control group that was vaccinated with tetanus toxoid. Thus the results of this trial up to

this date are that 12 cases of group A CSM occurred among the 62 054 children in the control group whereas no case occurred among the 62 295 children in the vaccine group. The difference is highly significant ( $P < 0.0005$ ).