Bull. Org. mond. Santé
Bull. Wld Hlth Org. } 1967, **37**, 703-727

A Controlled Field Trial of the Effectiveness of Cholera and Cholera El Tor Vaccines in the Philippines*

J. C. AZURIN, A. CRUZ, T. P. PESIGAN, M. ALVERO, T. CAMENA, R. SUPLIDO, L. LEDESMA & C. Z. GOMEZ

A controlled field trial on some 584 000 people in an endemic cholera El Tor area in the Philippines demonstrated that cholera vaccines gave moderate protection of short duration. Injection of a single dose of vaccine prepared from either Vibrio cholerae *or El Tor vibrios gave over 50 % protection for the first 2 months. The immunity conferred by the* V. cholerae *vaccine declined rapidly after 3 to 4 months. The effectiveness of the El Tor vaccine continued for 6 months. An oil-adjuvant vaccine prepared from* V. cholerae *conferred an equally high degree of protection for a longer period of time, but, owing to severe vaccination reactions, its use could not be recommended.*

Since Ferran (1885) introduced the first cholera vaccine 80 years ago, no evaluation of it had been made in a strictly controlled field trial (Cvjetanovic, 1965). Collaborative studies on the efficacy of cholera vaccine were initiated in 1964 following an agreement between the Government of the Philippines, the Government of Japan, and the World Health Organization. The controlled field study in the Philippines was planned by a joint Philippines–Japan–WHO committee and carried out to obtain new information on the efficacy of vaccines prepared from *Vibrio cholerae* and El Tor vibrio against infection with cholera El Tor.

Oil-adjuvant classical cholera vaccine developed and produced in Japan, and El Tor vaccine and classical cholera vaccine produced in the Philippines by methods in current use, were studied in a controlled trial on 584 026 persons. Funds, equipment and personnel for the study were provided by the two governments and the World Health Organization. A preliminary report on this study has been published earlier by the Philippines Cholera Committee (1965).

OBJECTIVES OF THE TIRAL

The objectives of the trials were:

(*a*) to assess and compare the protective power of classical *V. cholerae* vaccine, El Tor vibrio vaccine and oil-adjuvant classical *V. cholerae* vaccine against clinical attacks of cholera El Tor;

(*b*) to assess the protective power of the vaccines against asymptomatic infection with El Tor vibrios and the carrier stage;

(*c*) to assess the degree and duration of protection (if any); and

(*d*) to study the reaction of vaccinated people to each vaccine.

The study was organized as a strictly controlled field trial and the results obtained were analysed by the Joint Philippines–Japan–WHO Cholera Committee.

The effectiveness of each vaccine was evaluated by comparing the morbidity and carrier rates due to cholera El Tor among those vaccinated.

The group vaccinated with cholera vaccines and the group vaccinated with the control vaccine were similar with respect to age, sex, occupation, economic status and hygienic standards. This similarity was achieved by a random allocation of vaccines to the population.

The cholera vaccines used were demonstrated to be safe, and they met the potency tests described by the WHO Study Group on Requirements for... Cholera Vaccine (1959).

In order to avoid bias on the part of the observers and the observed, the cholera vaccines and the control vaccine were indicated by code letters. Cases

* From the Department of Health, Republic of the Philippines.

of cholera were thoroughly examined bacteriologically to confirm the diagnosis.

In order to satisfy ethical principes and to ensure public acceptance and co-operation, efforts were made to provide maximum health care for the population in the trial area.

The above and other relevant principles were applied when the protocol for the study was prepared.

DESIGN OF THE STUDY

General principles

Evaluation indicator. From the outset, it was anticipated that the number of deaths among bacteriologically confirmed cases in the three groups immunized with the different cholera vaccines would not reveal statistically significant differences from deaths among the control group. It was also an accepted fact that data reported by hospitals in the study area on the severity of cases would be inaccurate and therefore would not provide a good basis for evaluation. Thus, the best indicator was the number of bacteriologically confirmed cases and carriers.

Methods. To avoid bias, the principles of controlled field trials were strictly observed throughout the trial: a placebo (typhoid vaccine) and the double-blind test method were used, and the vaccines and placebo were labelled with code letters.

For the same reason, random allocation of vaccines was practised, applying a 4-item random allocation table following the Latin square pattern throughout the vaccination campaign. A provisional

TABLE 1

DISTRIBUTION OF PERSONS IN THE 4 VACCINE GROUPS ACCORDING TO AGE, IN A 1% SAMPLE OF THE VACCINATED POPULATION, 6 JUNE 1964 TO 5 DECEMBER 1965 [a]

Age (years)	Classical cholera vaccine		El Tor vaccine		Oil-adjuvant vaccine		Control vaccine		Total	
	No.	%	No.	%	No.	%	No.	%	No.	%
<1	21	1.44	16	1.08	17	1.18	16	1.09	70	1.20
1	46	3.16	58	3.92	43	2.99	54	3.68	201	3.44
2	52	3.57	53	3.58	40	2.79	59	4.02	204	3.49
3	66	4.54	68	4.59	66	4.60	55	3.75	255	4.37
4	56	3.85	71	4.79	63	4.39	62	4.22	252	4.32
5- 9	315	21.65	317	21.40	283	19.71	336	22.89	1 251	21.42
10-14	239	16.43	243	16.41	236	16.43	213	14.51	931	15.94
15-19	136	9.35	146	9.86	146	10.17	134	9.13	562	9.62
20-24	128	8.80	115	7.77	115	8.01	128	8.72	486	8.32
25-29	76	5.22	90	6.08	91	6.34	119	8.11	376	6.44
30-34	76	5.22	74	5.00	82	5.71	70	4.77	302	5.17
35-39	49	3.37	52	3.51	67	4.67	61	4.16	229	3.92
40-44	52	3.57	55	3.71	50	3.48	42	2.86	199	3.41
45-49	43	2.96	36	2.43	50	3.48	35	2.38	164	2.81
50-54	40	2.75	44	2.97	38	2.65	37	2.52	159	2.72
55-59	18	1.24	15	1.01	14	0.97	16	1.09	63	1.08
60-64	23	1.58	14	0.94	14	0.97	10	0.68	61	1.04
65-69	5	0.27	2	0.14	7	0.49	7	0.48	20	0.34
≥70	15	1.03	12	0.81	14	0.97	14	0.95	55	0.94
Total	1 455	100.00	1 481	100.00	1 436	100.00	1 468	100.00	5 840	99.99

[a] The estimated numbers of persons in the various groups in the entire vaccinated population may be obtained by multiplying the figures in the table by 100.

test on a 1% sample, analysing 5840 immunization cards selected at random, proved that random allocation of the 4 vaccines was successfully achieved: the 4 vaccinated groups were found to be comparable with regard to age, sex, geographical and occupational distributions, and history of infections and vaccinations (Tables 1 to 6).

Selection of trial area

Negros Occidental Province was chosen for the trial for the following reasons.

(1) According to data on the 3 severest epidemics of classical cholera and cholera El Tor in the Philippines in 1919, 1940 and 1961-62, Negros Occidental is one of the areas of highest risk.

(2) Except for some seasonal migration among sugar plantation workers, the general population is almost entirely settled.

(3) Field-study facilities were available. The former Maternity and Children's Hospital building in Bacolod City, recently vacated, was designated as the headquarters. It had ample space to house the epidemiological, statistical, and administrative offices; to store and file all 584 000 immunization cards; to house the bacteriological laboratories and media kitchen; and even to furnish project personnel with fairly comfortable living-quarters. Apart from the workers employed full-time on the project, personnel were provided by the Provincial Health Office and the City Health Department in Bacolod City, and the rural health units in the municipalities. In addition, when needed, some personnel of Regional Health Office No. 5 in Iloilo City (Panay Island) were assigned to the project. There were also a number of private and public hospitals throughout the province.

TABLE 2

DISTRIBUTION OF PERSONS IN THE 4 VACCINE GROUPS ACCORDING TO AGE AND SEX, IN A 1% SAMPLE OF THE VACCINATED POPULATION, 6 JUNE 1964 TO 5 DECEMBER 1965 [a]

Age (years)	Males								Females							
	Classical cholera vaccine		El Tor vaccine		Oil-adjuvant vaccine		Control vaccine		Classical cholera vaccine		El Tor vaccine		Oil-adjuvant vaccine		Control vaccine	
	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
0-4	115	15.88	134	17.54	113	15.83	111	15.61	126	17.24	132	18.41	116	16.30	135	17.83
5-9	153	21.13	167	21.86	143	20.03	150	21.10	162	22.16	150	20.92	140	19.34	186	24.57
10-14	108	14.92	131	17.15	112	15.69	104	14.63	131	17.92	112	15.62	124	17.13	109	14.40
15-19	68	9.39	75	9.82	64	8.96	64	9.00	68	9.30	71	9.90	82	11.33	70	9.25
20-24	81	11.19	52	6.81	63	8.82	63	8.86	47	6.43	63	8.79	52	7.18	65	8.59
25-29	42	5.80	42	5.50	48	6.72	61	8.58	34	4.65	48	6.69	43	5.94	58	7.66
30-34	29	4.01	41	5.37	44	6.16	29	4.08	47	6.43	33	4.60	38	5.25	41	5.42
35-39	27	3.73	29	3.80	34	4.76	29	4.08	22	3.01	23	3.21	33	4.56	32	4.23
40-44	20	2.76	27	3.53	26	3.64	23	3.23	32	4.38	28	3.91	24	3.31	19	2.51
45-49	21	2.90	14	1.83	26	3.64	22	3.09	22	3.01	22	3.07	24	3.31	13	1.72
50-54	22	3.04	27	3.53	18	2.52	23	3.23	18	2.46	17	2.37	20	2.76	14	1.85
55-59	12	1.66	10	1.31	7	0.98	11	1.55	6	0.82	5	0.70	7	0.97	5	0.66
60-64	16	2.21	8	1.05	6	0.84	6	0.84	7	0.96	6	0.84	8	1.10	4	0.53
65-69	0	0	2	0.26	4	0.56	6	0.84	4	0.55	0	0	3	0.41	1	0.13
≥70	10	1.38	5	0.65	6	0.84	9	1.27	5	0.68	7	0.98	8	1.10	5	0.66
Total	724	100.00	764	100.01	714	99.99	711	99.99	731	100.00	717	100.01	722	99.99	757	100.01

[a] The estimated numbers of persons in the various groups in the entire vaccinated population may be obtained by multiplying he figures in the table by 100.

(4) The experience of the epidemiological teams in Negros Occidental in 1962 indicated that the people of the province would submit to vaccination and surveillance.

(5) The inhabitants, settled mainly in the coastal regions, were easily accessible to project workers (see Fig. 1).

Determination of size of study population

The annual incidence of cholera El Tor in Negros Occidental is estimated to be 0.5 per 1000, on the basis of data from the 1962 epidemic. The endemicity of cholera El Tor in the Philippines is of the fluctuating type, unlike the Asiatic cholera encountered in the Ganges-Brahmaputra delta. The incidence of

FIG. 1
PROVINCE OF NEGROS OCCIDENTAL, WITH THE MUNICIPALITIES INVOLVED IN THE FIELD TRIAL

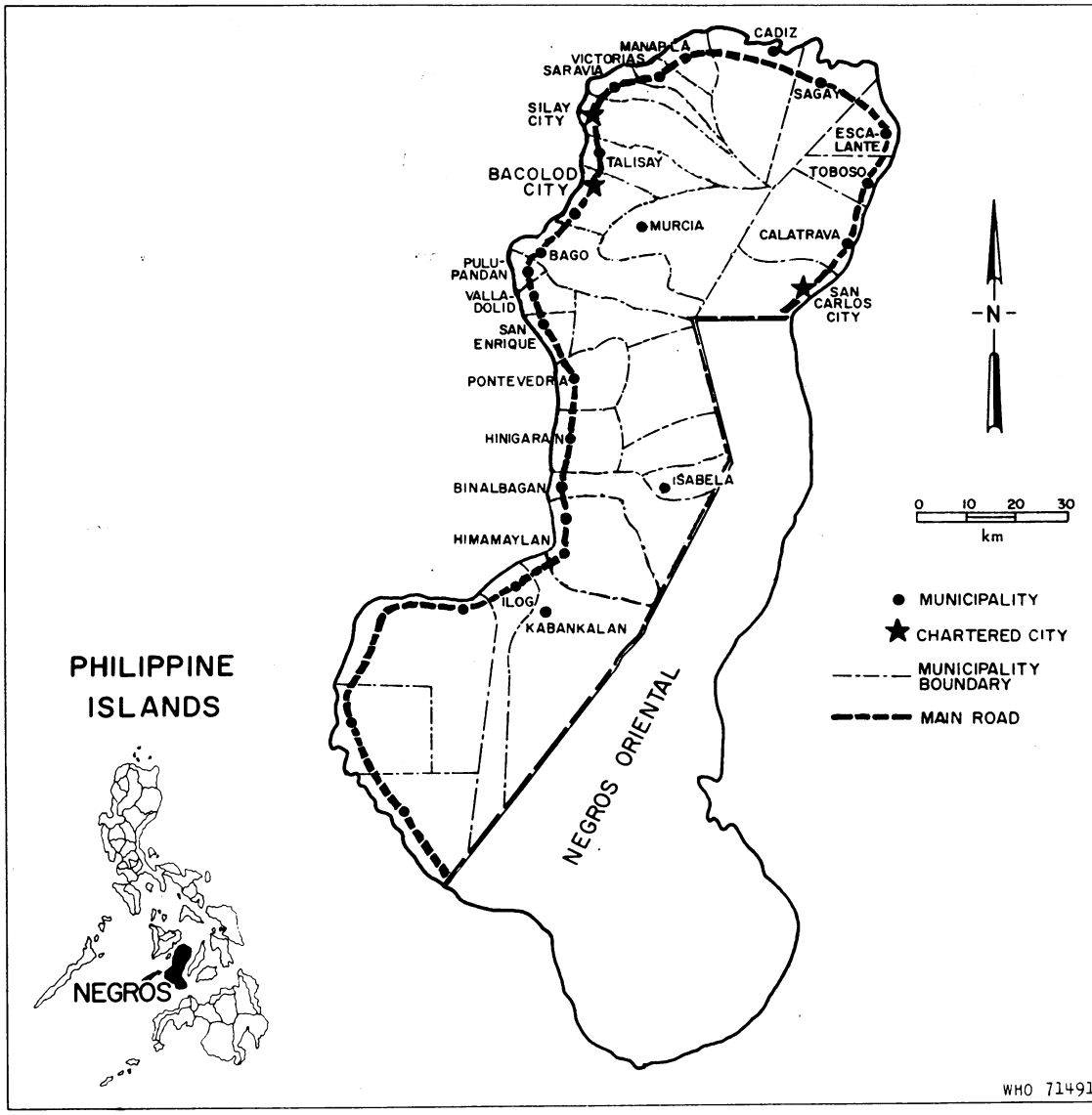

TABLE 3

DISTRIBUTION OF PERSONS IN THE 4 VACCINE GROUPS ACCORDING TO LOCALITY, IN A 1% SAMPLE OF THE VACCINATED POPULATION, 6 JUNE 1964 TO 5 DECEMBER 1965[a]

Locality	Classical cholera vaccine		El Tor vaccine		Oil-adjuvant vaccine		Control vaccine		Total	
	No.	%	No.	%	No.	%	No.	%	No.	%
Bago	78	5.36	102	6.89	96	6.69	118	8.04	394	6.75
Binalbagan	42	2.89	46	3.11	38	2.65	41	2.79	167	2.86
Cadiz	94	6.46	99	6.68	103	7.17	96	6.54	392	6.71
Escalante	88	6.05	72	4.86	77	5.36	68	4.63	305	5.22
Hinigaran	44	3.02	70	4.73	53	3.69	61	4.16	228	3.90
Manapla	39	2.68	35	2.36	33	2.30	45	3.07	152	2.60
Murcia	44	3.02	31	2.09	42	2.92	30	2.04	147	2.52
Pontevedra	28	1.92	47	3.17	45	3.13	35	2.38	155	2.65
Pulupandan	26	1.79	31	2.09	24	1.67	32	2.18	113	1.93
Sagay	129	8.87	108	7.29	93	6.48	102	6.95	432	7.40
San Enrique	18	1.24	15	1.01	14	0.97	21	1.43	68	1.16
Saravia	45	3.09	46	3.11	52	3.62	42	2.86	185	3.17
Talisay	69	4.74	62	4.19	81	5.64	61	4.16	273	4.67
Valladolid	23	1.58	20	1.35	25	1.74	24	1.63	92	1.58
Victorias	47	3.23	53	3.58	51	3.55	61	4.16	212	3.63
Bacolod	227	15.60	247	16.68	218	15.18	243	16.55	935	16.01
Silay	81	5.57	85	5.74	86	5.99	77	5.25	329	5.63
Toboso	27	1.86	35	2.36	28	1.95	30	2.04	120	2.05
Calatrava	60	4.12	52	3.51	48	3.34	65	4.43	225	3.85
San Carlos	95	6.53	84	5.67	74	5.15	90	6.13	343	5.87
Himamaylan	50	3.44	49	3.31	70	4.87	41	2.79	210	3.60
Kabankalan	42	2.89	38	2.57	38	2.65	31	2.11	149	2.55
Ilog	15	1.03	18	1.22	11	0.77	15	1.02	59	1.01
Isabela	44	3.02	36	2.43	36	2.51	39	2.66	155	2.65
Total	1 455	100.00	1 481	100.00	1 436	100.00	1 468	100.00	5 840	99.97

[a] The estimated numbers of persons in the various groups in the entire vaccinated population may be obtained by multiplying the figures in the table by 100.

endemic cholera El Tor in Negros Occidental was very high in the summer of 1962 (Joseph et al., 1965) but was very low in 1963; it was therefore expected that there would be a rather high cholera wave in the summer of 1964. Besides encouraging case-reporting, the plan was to detect as many cholera cases as possible by enlisting the co-operation of barrio officials in reporting diarrhoeal patients. Taking the above factors into consideration, the incidence of 0.5 per 1000 was regarded as a safe working estimate. As a matter of fact, from the start of surveillance work in July 1964 until 5 December 1965, the incidence rate of cholera El Tor in the placebo vaccine group had risen to more than 0.8 per 1000.

In order to achieve statistically significant differences between the vaccinated and control groups, even if the effectiveness of the vaccine were as low as 33%, it was decided that the vaccines would be tried on a minimum of 130 000 people per vaccine

TABLE 4

DISTRIBUTION OF PERSONS IN THE 4 VACCINE GROUPS ACCORDING TO OCCUPATION, IN A 1% SAMPLE OF THE VACCINATED POPULATION, 6 JUNE 1964 TO 5 DECEMBER 1965 [a]

Occupation	Classical cholera vaccine		El Tor vaccine		Oil-adjuvant vaccine		Control vaccine		Total	
	No.	%	No.	%	No.	%	No.	%	No.	%
Administrative, executive	3	0.2	1	0.1	2	0.1	1	0.1	7	0.1
Professional, technical	11	0.7	114	0.8	11	0.8	13	0.9	49	0.8
Clerical, sales	33	2.3	38	2.6	39	2.7	39	2.7	149	2.6
Subordinate technical	261	17.9	256	17.3	280	19.5	281	19.1	1 078	18.5
Service occupations	241	16.6	229	15.5	256	17.8	229	15.6	955	16.3
Manual workers	906	62.3	943	63.7	848	59.1	905	61.6	3 602	61.7
Total	1 455	100.0	1 481	100.0	1 436	100.0	1 468	100.0	5 840	100.0

[a] The estimated numbers of persons in the various groups in the entire vaccinated population may be obtained by multiplying the figures in the table by 100.

group or a total of 520 000 for the 4 vaccine groups (Nobechi, 1965). In fact, a total of 584 026 persons were vaccinated—about 146 000 persons per vaccine. Thus, evaluation was facilitated and more statistically significant results were obtained. For instance, the classical cholera vaccine, although only 26% effective, revealed a significant difference (P=5%) compared with the placebo vaccine.

POPULATION CHARACTERISTICS

The investigation cards used in the vaccine studies had been designed for IBM machine analysis. However, such analysis was not carried out. Instead, the characteristics of the study population, upon which various observations in the present studies are based, were derived from the result of a test of a random 1% sample of the 584 000 immunization cards, as stated above. This was done primarily to ascertain whether random allocation of the 4 vaccines was satisfactorily achieved. The numbers vaccinated with classical cholera, El Tor, oil-adjuvant and control vaccines were estimated to be 145 500, 148 000, 143 800 and 146 800 respectively, as shown in Table 1.

The geographical and occupational distributions among the 4 vaccine groups are likewise given in Tables 3 and 4.

As stated above, the population characteristics of the 4 vaccine groups were found to be comparable, not only with respect to age, sex, geographical and occupational distribution, but also in relation to histories of infection and vaccination (Tables 5 and 6).

VACCINES

The following vaccines were used in the controlled field trial.

(*a*) Classical cholera vaccine prepared in the Philippines according to the method adopted by the Bureau of Research and Laboratories in Manila. This vaccine was made from *V. cholerae*, strains Inaba 35A-3 and Ogawa 41, and met the requirements established by the WHO Study Group on Requirements for . . . Cholera Vaccine (1959).

(*b*) Cholera El Tor fluid vaccine prepared in the same laboratory from lyophilized cultures of local El Tor strains—Ogawa 1418 and Inaba 6973.

(*c*) Classical cholera oil-adjuvant vaccine prepared according to the method developed by H. Ogonuki of the Chiba Serum Institute, Japan, using *V. cholerae* strains Inaba 35A-3 and Ogawa 41.

(*d*) Monovalent typhoid vaccine, which was used as a control, prepared by the Bureau of Research and Laboratories, Manila. Full details of the first 3 vaccines are given by Pesigan et al. (1967) and Ogonuki et al. (1967).[1]

[1] See the papers on p. 816 and 729 of this issue.

TABLE 5

PREVIOUS HISTORY OF INFECTIOUS DISEASES AMONG VACCINEES, IN A 1% SAMPLE OF THE VACCINATED POPULATION, 6 JUNE 1964 TO 5 DECEMBER 1965 [a]

Vaccine	No. in sample	Previous history of infectious diseases							
		Cholera				Typhoid			
		Positive		Negative		Positive		Negative	
		No.	%	No.	%	No.	%	No.	%
Classical cholera	1 455	6	0.4	1 449	99.6	9	0.6	1 446	99.4
El Tor	1 481	9	0.6	1 472	99.4	5	0.3	1 476	99.7
Oil-adjuvant	1 436	9	0.6	1 427	99.4	7	0.5	1 429	99.5
Control	1 468	10	0.7	1 458	99.3	8	0.5	1 460	99.5
Total	5 840	34	0.6	5 806	99.4	29	0.5	5 811	99.5

[a] The estimated numbers of persons in the various groups in the entire vaccinated population may be obtained by multiplying the figures in the table by 100.

DOSAGE

The cholera vaccines were administered in a single dose, according to the following scheme.

Age (years)	*Dose (ml)*
Classical and El Tor vaccines	
0-4	0.25
5-9	0.5
10 +	1.0
Classical oil-adjuvant vaccine	
0-4	0.05
5-9	0.1
10 +	0.2

The oil-adjuvant vaccine contained 2×10^{10} organisms per ml. Vaccines were bottled in 50-ml containers marked with appropriate code letters. All vaccinations were given subcutaneously in the upper arm.

MASS VACCINATION CAMPAIGN

In anticipation of a rise in the incidence of cholera, intensive preparation for a mass vaccination campaign began during the first week of May. Vaccinators, recorders and supervisors were recruited and trained. Instructions for field teams were distributed

TABLE 6

PREVIOUS HISTORY OF VACCINATION AMONG VACCINEES, IN A 1% SAMPLE OF THE VACCINATED POPULATION, 6 JUNE 1964 TO 5 DECEMBER 1965 [a]

Vaccine administered during field trial	No. in sample	Previously vaccinated against cholera				Previously vaccinated against smallpox			
		Positive		Negative		Positive		Negative	
		No.	%	No.	%	No.	%	No.	%
Classical cholera	1 455	1 156	79.5	299	20.5	984	67.6	471	32.4
El Tor	1 481	1 192	80.5	289	19.5	970	65.5	511	34.5
Oil-adjuvant	1 436	1 173	81.7	263	18.3	948	66.0	488	34.0
Control	1 468	1 177	80.2	291	19.8	971	66.1	497	33.9
Total	5 840	4 698	80.4	1 142	19.6	3 873	66.3	1 967	33.7

[a] The estimated numbers of persons in the various groups in the entire vaccinated population may be obtained by multiplying the figures in the table by 100.

and used as a guide in the training. Each vaccination team was composed of 1 recorder and 1 vaccinator. The recorder filled in the cholera immunization cards and the random allocation cards. The vaccinator administered the vaccines as indicated by the recorder.

The study area was divided into 3 sectors. A field supervisor was assigned to each sector, each supervisor having in his charge 54 teams. A total of 162 teams were thus sent to the field.

The vaccination campaign was conducted in public places such as markets and schools, in places of work such as sugar mills and plantations, and on a house-to-house basis. Vaccination was, however, administered only to volunteers. Random allocation of vaccines to volunteers was achieved by a random ordering of vaccine code letters, strictly adhered to.

The vaccination campaign was initiated on 18 May 1964 and terminated on 4 July 1964, the duration being about 7 weeks. Records show that 584 026 persons were vaccinated.

When the Joint Philippines–Japan–WHO Cholera Committee drew up the protocol for this vaccine trial, the population in the trial area was estimated to be 892 274. However, during the actual vaccination campaign, certain municipalities were incorporated into the trial area, which brought the total estimated population up to 1 339 870. This is indicated in Table 7. Considering geographical accessibility and other factors, a target population of 937 909 was chosen for vaccination. However, the actual number of vaccinations was only 584 026, as shown in Table 7. Thus, the average proportion of vaccinees among the target population was 43.6%; the highest proportion was 67.9% in Bacolod City, and the lowest was 21.0% in Ilog.

SURVEILLANCE

Immediately after the vaccination campaign, intensive surveillance was undertaken to detect cholera El Tor cases among the vaccinated groups.

Surveillance was performed by 3 epidemiologists, 8 nurse supervisors and 26 epidemiological aides (trained midwives). The entire study area was divided into 6 sectors to facilitate the work. Each surveillance sector was under the supervision of a nurse supervisor, with the other 2 nurse supervisors providing co-ordination. Each municipality was assigned an epidemiological aide whose only task was to look for cases. Assistance was given by 263 staff members of local health units within the study area. In addition, councillors and barrio captains were enjoined to report diarrhoeal cases occurring in their communities.

Diarrhoeal patients reporting to rural health units, local private physicians, barrio captains, pharmaceutical stores and schools constituted suspected cases. This number was supplemented by diarrhoeal cases revealed by a house-to-house inquiry. Upon being notified of such cases, the epidemiological aides in the municipalities concerned made calls in the patients' homes. Rectal swabs from the persons with diarrhoea were collected (once), and the material was inoculated into alkaline peptone water. These specimens were taken to the project laboratory in Bacolod for bacteriological examination. If the cultures proved to be positive, the epidemiologists continued their investigations and searched for secondary cases or carriers. These strains were all sent to Tokyo and Manila for further studies, and likewise all clinical and epidemiological records were scrutinized by a competent group of bacteriologists, clinicians and epidemiologists from Philippines, Japan and WHO staff to avoid bias. All household contacts of confirmed cases were subjected to rectal swabbing. Appropriate records were made of all cases and carriers. The rather high morbidity rate of cholera El Tor in the trial area in 1964 may be attributed to such special and intensive surveillance.

RESULTS

Incidence and incidence rate

Case rate among vaccinated groups. The aggregate numbers of cholera El Tor cases which occurred during the surveillance period, divided into 3 periods of 6 months each, are given in Table 8. All these cases were confirmed bacteriologically. A total of 499 cases of cholera El Tor were found within the 4 vaccine groups.

The incidence rates in the different vaccine groups are also indicated in Table 8. It may be observed that in the first 6 months (6 June 1964 to 5 December 1964) the control group showed the highest incidence rate, 87.9 per 100 000 population, the second highest being 64.6 among those who received the classical cholera vaccine. The El Tor vaccine group had an incidence rate of 48.6 while the oil-adjuvant vaccine group had the lowest incidence rate, 39.7.

Monthly distribution of cases. The monthly distribution of cholera cases is shown in Table 9.

TABLE 7

VACCINATION DATA IN NEGROS OCCIDENTAL IN 1964

Sector	Locality	Estimated population	Target population	Vaccinated population		No. of teams	Team days	Date begun	Date finished
				No.	%				
I	San Carlos [a]	149 220	104 454	34 258	23.0	51	393	27 June	4 July
	Calatrava [a]	75 686	52 980	22 492	29.7	35	167	22 June	26 June
	Toboso [a]	41 989	29 393	12 024	28.6	26	102	22 June	26 June
	Total	266 895	186 827	68 774	25.8	—	662	22 June	4 July
II	Escalante	68 977	48 284	30 479	44.2	35	311	18 May	30 May
	Sagay	82 118	57 482	43 224	52.6	30	383	18 May	1 June
	Cadiz	102 027	71 419	39 167	38.4	35	364	18 May	30 May
	Total	253 122	177 185	112 870	44.6	—	1 058	18 May	1 June
III	Manapla	54 064	37 845	15 144	28.0	30	118	2 June	5 June
	Victorias	40 107	28 075	21 208	52.9	27	158	1 June	6 June
	Saravia	36 851	25 795	18 466	50.1	36	177	29 May	4 June
	Silay	69 500	48 650	32 871	47.3	35	305	5 June	15 June
	Total	200 522	140 365	87 689	43.7	—	758	29 May	15 June
IV	Talisay	53 571	37 500	27 270	50.9	29	251	6 June	15 June
	Bacolod	137 740	96 418	93 561	67.9	137	861	30 May	3 July
	Murcia	26 753	18 727	14 718	55.0	27	131	8 June	12 June
	Total	218 064	152 645	135 549	62.2	—	1 243	30 May	3 July
V	Bago	68 054	47 638	39 414	57.9	26	348	27 May	17 June
	Pulupandan	17 752	12 427	11 313	63.7	29	105	26 June	29 June
	Valladolic	16 959	11 871	9 194	54.2	28	102	26 June	29 June
	San Enrique	12 155	8 509	6 856	56.4	15	59	22 June	25 June
	Pontevedra	26 316	18 421	15 453	58.7	36	146	22 June	25 June
	Hinigaran	43 369	30 358	22 864	52.7	25	158	15 June	23 June
	Total	184 605	129 224	105 094	56.9	—	918	27 May	29 June
VI	Binalbagan	35 973	25 181	16 765	46.6	47	132	12 June	26 June
	Himamaylan [a]	48 659	34 060	20 991	43.1	52	181	24 June	1 July
	Isabela [a]	34 117	23 882	15 433	45.2	51	203	30 June	3 July
	Kabankalan [a]	69 578	48 705	14 912	21.4	43	190	29 June	4 July
	Ilog [a]	28 336	19 835	5 949	21.0	32	66	1 July	4 July
	Total	216 662	151 663	74 050	34.2	—	772	12 June	4 July
	Grand total	1 339 870	937 909	584 026	43.6		5 411	18 May	4 July

[a] These municipalities were added in late June 1964 to the original list designated in the Protocol, because the number of vaccinees had not come up to 520 000 as intended.

TABLE 8

CHOLERA EL TOR AMONG VACCINE GROUPS, 6 JUNE 1964 TO 5 DECEMBER 1965

Vaccine	No. vaccinated	Incidence							
		6 June to 5 Dec. 1964		6 Dec. 1964 to 5 June 1965		6 June to 5 Dec. 1965		Total	
		No.	Rate per 100 000	No.	Rate per 100 000	No.	Rate per 100 000	No.	Rate per 100 000
Classical cholera	145 500	94	64.6	33	22.7	4	2.7	131	90.0
El Tor	148 100	72	48.6	34	23.0	7	4.7	113	76.3
Oil-adjuvant	143 600	57	39.7	20	13.9	7	4.9	84	58.5
Control	146 800	129	87.9	38	25.9	4	2.7	171	116.5
Total	584 000	352	60.3	125	21.4	22	3.8	499	85.4

TABLE 9

MONTHLY DISTRIBUTION OF CHOLERA CASES, 6 JUNE 1964 TO 5 DECEMBER 1965

Period	Classical cholera vaccine group	El Tor vaccine group	Oil-adiuvant vaccine group	Control vaccine group	Total
6-30 June 1964	1	0	0	0	1
1-31 July	8	4	13	23	48
1-31 August	9	11	8	24	52
1-30 September	16	14	12	18	60
1-31 October	24	21	5	25	75
1-30 November	27	20	13	32	92
1-31 December	25	18	18	30	91
1-31 January 1965	15	16	6	13	50
1-28 February	2	2	2	2	8
1-31 March	0	0	0	0	0
1-30 April	0	0	0	0	0
1-31 May	0	0	0	0	0
1-30 June	0	1	0	0	1
1-31 July	0	0	0	1	1
1-31 August	0	0	0	0	0
1-30 September	0	0	0	0	0
1-31 September	0	1	4	1	6
1-30 November	4	5	3	2	14
1-5 December	0	0	0	0	0
Total	131	113	84	171	499

The cases began to occur in June, which is the start of the rainy season. The peak of the endemic period is seen in October, November and December.

Distribution of cases between vaccination and onset of illness. Tables 10 and 11 show, in general by 15-day and 30-day intervals, the occurrence of confirmed cholera cases in each vaccine group reckoned from the time of vaccination to the onset of illness. These two tables may indicate the period of immunity more clearly than Table 9.

Table 12 shows the cumulative distribution of cholera cases from vaccination to onset of illness by 15-day intervals for the first 6 months, and by 30-day intervals for the following 12 months. The effectiveness of the various vaccines calculated from the data of Table 12 is shown in Table 13.

Table 14 gives the incidence rates by vaccine group and by time (in days) from vaccination to onset of illness, together with the statistical significance of the differences in incidence rates between the various vaccine groups and the control group.

During the first 2 months after vaccination, statistically significant differences were obtained between the various vaccine groups and the control group, while the differences in incidence rates among the cholera vaccine groups were not statistically significant.

From 61 to 120 days after vaccination, the differences in incidence rates between the El Tor vaccine group and the control group, and also between the oil-adjuvant vaccine group and the control group, were statistically significant. The differences between the cholera vaccine group and the control group were not statistically significant.

Five to 6 months after vaccination, the differences between the El Tor vaccine and the control group, and between the oil-adjuvant vaccine and the control group, were still statistically significant. The difference between the cholera and oil-adjuvant vaccine groups was also statistically significant. It is pertinent to mention that the difference in incidence rates between the cholera and oil-adjuvant vaccine groups was statistically significant only during this 2-month

TABLE 10

CONFIRMED CHOLERA CASES BY 15-DAY PERIODS FROM VACCINATION TO ONSET OF ILLNESS, JUNE-DECEMBER 1964 [a]

Days from vaccination to illness	No. of cholera cases				Total
	Classical cholera vaccine group	El Tor vaccine group	Oil-adjuvant vaccine group	Control vaccine group	
0-7	0	0	0	0	0
8-15	1	0	0	3	4
16-22	5	1	2	2	10
23-30	0	2	2	7 (1)	11 (1)
31-45	2	3	9	8	22
46-60	5	7	4	15 (1)	31 (1)
61-75	6	7	4	8	25
76-90	7 (1)	4	6 (1)	13	30 (2)
91-105	9 (1)	7	6	13 (1)	35 (2)
106-120	8	7 (1)	3	10	28 (1)
121-135	10 (1)	7 (1)	1	4 (1)	22 (3)
136-150	10	7 (2)	6	13 (1)	36 (3)
151-165	14 (5)	14 (1)	2	22 (3)	52 (9)
166-180	14 (1)	3	10 (1)	16 (3)	43 (5)
Total	91 (9)	69 (5)	55 (2)	134 (11)	349 (27)

[a] The numbers in parentheses indicate fatal cases.

TABLE 11
CONFIRMED CHOLERA CASES BY 30-DAY PERIODS FROM VACCINATION TO ONSET OF ILLNESS, 6 JUNE 1964 TO 5 DECEMBER 1965

Days from vaccination to illness	No. of cholera cases				Total
	Classical cholera vaccine group	El Tor vaccine group	Oil-adjuvant vaccine group	Control vaccine group	
8-30	6	3	4	12 (1)	25 (1)
31-60	7	10	13	23 (1)	53 (1)
61-90	13 (1)	11	10 (1)	21	55 (2)
91-120	17 (1)	14 (1)	9	23 (1)	63 (3)
121-150	20 (1)	14 (3)	7	17 (2)	58 (6)
151-180	28 (6)	17 (1)	12 (1)	38 (6)	95 (14)
181-210	19	23	16	22 (2)	80 (2)
211-240	10 (1)	7 (2)	3	8	28 (3)
241-270	7 (1)	5 (1)	3	3	18 (2)
271-300	0	2	0	0	2
301-330	0	0	0	0	0
331-360	0	0	0	0	0
361-390	0	1	0	1	2
391-420	0	0	0	0	0
421-450	0	0	0	0	0
451-480	0	0	0	0	0
481-510	1	1	3 (1)	1	6 (1)
511-540	3 (2)	5 (2)	4	2	14 (4)
Total	131 (13)	113 (10)	84 (3)	171 (13)	499 (39)

[a] The numbers in parentheses indicate fatal cases.

period. The differences between the El Tor vaccine and cholera vaccine groups, the cholera vaccine and control groups and the El Tor and oil-adjuvant vaccine groups were not statistically significant.

From 7 to 8 months after vaccination, there were no statistically significant differences between any of the vaccine groups and the control group nor among the vaccine groups.

During the 9-to-10-month period (241–300 days), the number of cases among those given the cholera and El Tor vaccines was actually more than among those who received the control vaccine. The oil-adjuvant vaccine group had nearly the same incidence rate as the control group in this period.

Age distribution. As shown in Table 15, 51% of all cases occurred in children below 9 years of age. This is more or less indicative of the endemic pattern of the disease in the Philippines.

Table 16 gives the efficacy of 3 vaccines from 6 June 1964 to 5 December 1965, by age-groups. A high morbidity was found in the youngest age-group (0.5–9 years) and in the oldest age-group (25 years or more), with a lower morbidity in the middle group (10–24 years).

An interesting point made by these tables is that classical cholera vaccine did not protect the age-group 0.5–9 years. The El Tor and classical oil-adjuvant vaccines did protect this age-group to some extent, though not very impressively. The efficacies of these 3 vaccines in the middle age-group (10–24 years) were almost comparable, being 51%, 42% and 69% for classical, El Tor and oil-adjuvant vaccines respectively. Reasonable protection occurred

TABLE 12

CUMULATIVE DISTRIBUTION OF CHOLERA CASES, BY TIME INTERVAL FROM VACCINATION TO ONSET OF ILLNESS, 6 JUNE 1964 TO 5 DECEMBER 1965

Days from vaccination to illness	No. of cholera cases			
	Classical cholera vaccine group	El Tor vaccine group	Oil-adjuvant vaccine group	Control vaccine group
8-15	1	0	0	3
16-30	6	3	4	12
31-45	8	6	13	20
46-60	13	13	17	35
61-75	19	20	21	43
76-90	26	24	27	56
91-105	9	7	6	13
106-120	17	14	9	23
121-135	27	21	10	27
136-150	37	28	16	40
151-165	51	42	18	62
166-180	65	45	28	78
181-210	19	23	16	22
211-240	29	30	19	30
241-270	36	35	22	33
271-300	36	37	22	33
301-330	36	37	22	33
331-360	36	37	22	33
361-390	—	1	—	1
391-420	—	1	—	1
421-450	—	1	—	1
451-480	—	1	—	1
481-510	1	2	3	2
511-540	4	7	7	4

in the group older than 25 years, in spite of a high attack rate in the control group.

Thus, the above observations indicate that:

(*a*) in spite of a high attack rate in the oldest age-group, this group could be protected, to some extent, by immunization with the 3 vaccines;

(*b*) the youngest age-group could not be protected as effectively as the 2 older age-groups;

(*c*) since approximately 51% of the attacks occurred among those below 9 years of age, the lower average efficacy of the classical cholera vaccine, as compared with the other 2 vaccines during the 6-month observation periods, seems to be related to statement (*b*).

Sex distribution. Table 17 shows that both sexes were equally affected by the cholera El Tor infection: of the total number of cases, 49% occurred among males and 51% among females.

Seasonal and geographical distribution. The relative incidence rates in towns and cities are shown in Table 18. The first 10 months of the period covered by the study seem to show an unprecedentedly high incidence rate. The total number of cases from September to December 1964 is appreciably higher than during the same period in 1965. This might simply mean that 1964 was a peak year for this infection while 1965 was a low-incidence year, following the normal fluctuating endemic pattern.

In June 1964, which is the beginning of the rainy season, cholera El Tor cases began to appear in some parts of the province, most of them in the southern section. The predominance of cases south of Bacolod City continued until September when cases cropped up in the northern towns and cities. From October 1964 to February 1965, there were definitely more cases north of Bacolod than in the southern towns. Bacolod City itself had a steady incidence rate for the same period of time.

Following the seasonal pattern of the past few years, cases occurred in San Carlos City and in the northern towns of the province towards the end of the season of prevalence. Calatrava was free of cases until October 1964, Toboso until August and Victorias until September. The incidence in the coastal areas and the adjacent plains was higher than in the interior.

Table 19 shows that about 27% of the cases were discovered at home by a house-to-house search. This table also shows the proportions of hospital and home cases for the various vaccine groups.

Effectiveness of cholera vaccines

Monthly distribution of effectiveness of cholera vaccines. The classical cholera vaccine gave only 50% protection during the first month after vaccination, but 70% during the second month. This decreased to 38% in the third month and 26% in the fourth while during the fifth month no protective effect was noticeable (Table 13).

TABLE 13. EFFECTIVENESS OF CHOLERA VACCINES, BY TIME INTERVAL FROM VACCINATION TO ONSET OF ILLNESS, 6 JUNE 1964 TO 5 DECEMBER 1965 [a]

Days from vaccination to onset of illness	Classical cholera vaccine			El Tor vaccine			Oil-adjuvant vaccine			Control vaccine	
	No. of cases	Incidence rate per 100 000	Effectiveness (%)	No. of cases	Incidence rate per 100 000	Effectiveness (%)	No. of cases	Incidence rate per 100 000	Effectiveness (%)	No. of cases	Incidence rate per 100 000
0-30	6	4.1	50	3	2.0	76	4	2.8	66	12	8.2
31-60	7	4.8	69	10	6.8	57	13	9.1	42	23	15.7
61-90	13	8.9	38	11	7.4	48	10	7.0	51	21	14.3
91-120	17	11.7	26	14	9.5	40	9	6.3	60	23	15.7
121-150	20	13.7	—	14	9.5	18	7	4.9	58	17	11.6
151-180	28	19.2	26	17	11.5	56	12	8.4	68	38	25.9
0-60	13	8.9	63	13	8.8	63	17	11.8	50	35	23.8
61-120	30	20.6	31	25	16.9	44	19	13.2	56	44	30.0
121-180	48	33.0	12	31	20.9	44	19	13.2	65	55	37.5
181-240	29	19.9	3	30	20.3	1	19	13.2	35	30	20.4
241-300	7	4.8	—	7	4.7	—	3	2.1	0	3	2.0
301-360	0	0.0	—	0	0.0	—	0	0.0	—	0	0.0
0-180	91	62.5	32	69	46.6	49	55	38.3	58	134	91.3
181-360	36	24.7	—	37	25.0	—	22	15.3	32	33	22.5
361-540	4	2.7	0	7	4.7	—	7	4.9	—	4	2.7

[a] The effectiveness of the vaccines is expressed as the percentage reduction in the incidence rate in vaccinated groups as compared with the control group. A dash indicates that the incidence in the vaccinated group was greater than in the control group.

TABLE 14. INCIDENCE RATES (PER 100 000), BY VACCINE GROUP AND TIME INTERVAL BETWEEN VACCINATION AND ONSET OF ILLNESS, AND SIGNIFICANCE OF THE DIFFERENCE BETWEEN INCIDENCE RATES FOR VARIOUS VACCINE GROUPS

Vaccine group or groups	Time interval (days)				
	0-60	61-120	121-180	181-240	241-300
	Incidence rate (per 100 000)				
Classical cholera	8.93	20.62	32.99	19.93	4.81
El Tor	8.78	16.88	20.93	20.26	4.73
Oil-adjuvant	11.84	13.23	13.23	13.23	2.09
Control	23.84	29.97	37.47	20.44	2.04
	Student's *t* for difference between vaccine groups				
Control/Classical	3.15 [a]	1.59	0.64	0.10	−1.28 [b]
Control/El Tor	3.20 [a]	2.32 [a]	2.62 [a]	0.03	−1.26 [b]
Control/Oil-adjuvant	2.43 [a]	3.08 [a]	4.11 [a]	1.50	0.03
Classical/El Tor	0.04	0.74	1.99	0.06	0.03
Classical/Oil-adjuvant	0.77	1.53	3.50 [a]	1.40	1.25
El Tor/Oil-adjuvant	0.81	0.80	1.61	1.47	1.22

[a] Significant at 5 % level.
[b] No vaccine effect (more cases in vaccine group than in control group).

TABLE 15

CHOLERA CASES IN EACH VACCINE GROUP, BY AGE, 6 JUNE 1964 TO 5 DECEMBER 1965

Age-group (years)	Classical cholera vaccine group			El Tor vaccine group			Oil-adjuvant vaccine group			Control vaccine group			Total No. of cholera cases
	Percentage of total population	No. of cholera cases	Percentage of total cholera cases	Percentage of total population	No. of cholera cases	Percentage of total cholera cases	Percentage of total population	No. of cholera cases	Percentage of total cholera cases	Percentage of total population	No. of cholera cases	Percentage of total cholera cases	
0.5-1	1.4	7	5.3	1.1	3	2.6	1.2	4	4.8	1.1	1	0.6	15
1-4	15.1	34	26.0	16.9	42	37.2	14.8	20	23.8	15.7	45	26.3	141
5-9	21.7	31	23.7	21.4	16	14.2	19.7	21	25.0	22.9	29	17.0	97
10-14	16.4	9	6.9	16.4	10	8.8	16.4	3	3.6	14.5	11	6.4	33
15-24	18.2	6	4.6	17.6	7	6.2	18.2	6	7.1	17.8	17	10.0	36
25-34	10.4	11	8.4	11.1	15	13.3	12.1	8	9.5	12.9	22	12.8	56
35-44	7.0	16	12.2	7.2	6	5.3	8.1	11	13.0	7.0	14	8.2	47
45-54	5.7	11	8.4	5.4	5	4.4	6.1	4	4.8	4.9	9	5.3	29
55-64	2.8	4	3.0	2.0	6	5.3	1.9	4	4.8	1.8	13	7.6	27
⩾65	1.3	2	1.5	0.9	3	2.7	1.5	3	3.6	1.4	10	5.8	18
Total	100.0	131	100.0	100.0	113	100.0	100.0	84	100.0	100.0	171	100.0	499

The El Tor vaccine showed a high protective effect as early as the first month; the effect decreased gradually to 18% in the fifth month and then unexpectedly increased to 55% in the sixth month—owing to the abnormally high incidence among the control group in this month.

The oil-adjuvant vaccine gave immediate high protection which decreased slightly in the second month, but steadily increased in succeeding months, being still at a high level by the end of 6 months (Table 13).

The variation of the effectiveness of the various vaccines during the first 7 months after vaccination is shown in Fig. 2.

The effectiveness of the classical cholera vaccine approached 60% in the first month after the vaccination, but it declined rather sharply, and toward the end of the sixth month it was as low as 10%, while that of the El Tor vaccine reached about 70% in the first month, to decline to 15% towards the end of the sixth month. The effectiveness of the oil-adjuvant vaccine was nearly 60% in the first month, and about 50% towards the end of the sixth month, which suggested that the effectiveness might persist

FIG. 2

EFFECTIVENESS OF CHOLERA VACCINES FROM 1 TO 7 MONTHS AFTER VACCINATION

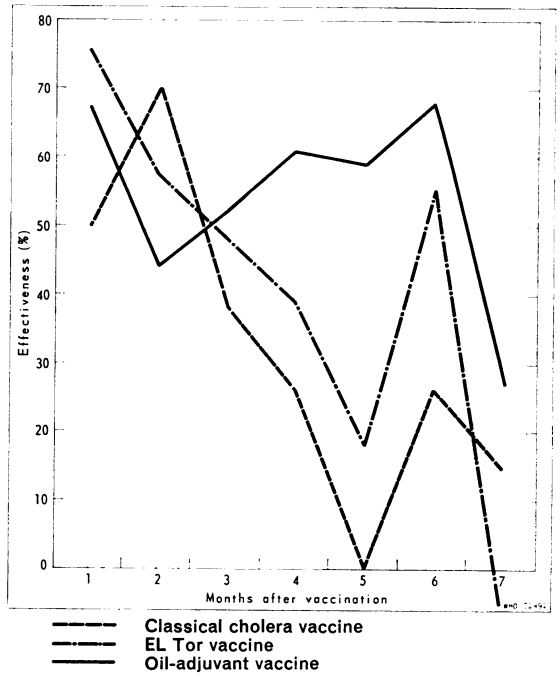

TABLE 16
EFFECTIVENESS OF THE CHOLERA VACCINES, 6 JUNE 1964 TO 5 DECEMBER 1965

Age-group (years)	Vaccine group			
	Classical cholera	El Tor	Oil-adjuvant	Control
	No. of cases			
0.5-9	72	61	45	75
10-24	15	17	9	28
⩾25	44	35	30	68
	No. of persons vaccinated			
0.5-9	55 600	58 300	51 400	58 200
10-24	50 300	50 400	49 700	47 500
⩾25	39 600	39 400	42 700	41 100
	Incidence rate (per 100 000)			
0.5-9	129	105	88	129
10-24	30	34	18	59
⩾25	111	89	70	165
	Effectiveness (%)			
0.5-9	0	19	32	—
10-24	49	42	70	—
⩾25	33	46	58	—

for a considerable period of time. However, because of the advent of the low-incidence season, the duration of immunity could not be tested accurately.

Mortality and fatality rates

Mortality. The number of deaths among the various cholera vaccine groups is shown in Table 20. A total of 39 deaths occurred among the population of 584 000, giving an average death rate of 6.7 per 100 000. The death rate among the oil-adjuvant group, 2.1 per 100 000, is significantly less than that in the control group.

Table 21 shows that deaths in each vaccine group, reckoned in 6-month periods from the date of vaccination, correspond closely to the number of cases occurring during the same period.

Fatality. The case-fatality rates among the vaccinated groups, shown in Table 22, follow the pattern of the morbidity rates. The rate for the oil-adjuvant vaccine group, 3.6%, is the only one to be significantly lower than that of 7.6% in the control group.

The age-specific distribution of deaths and fatality rates was compared among the 4 vaccine groups; the results obtained are shown in Table 23. The number of deaths and, accordingly, the fatality rates were decidedly higher in the lower age-group. Neither the classical cholera nor the El Tor vaccine seemed to be of any effect in preventing deaths from cholera El Tor in either age-group. The oil-adjuvant classical cholera vaccine, however, halved the fatality of cholera in the lower age-group; no conclusion could be drawn for the higher age-group.

TABLE 17
CONFIRMED CHOLERA CASES, BY SEX, 6 JUNE 1964 TO 5 DECEMBER 1965

Sex	Classical cholera vaccine group	El Tor vaccine group	Oil-adjuvant vaccine group	Control vaccine group	Total
Male	68	48	43	89	248
Female	63	65	41	82	251
Total	131	113	84	171	499

Severity

The duration of diarrhoea in confirmed cholera cases among the 4 vaccine groups is shown in Table 24. Fatal cases were not included among these figures. Analysis of this table will show that diarrhoea was present in all cases and was approximately of the same duration in the various cholera vaccine groups as in the control group.

An analysis of the symptoms of the 499 cases in the different cholera vaccine groups and the control group is shown in Table 25. In all of these cases diarrhoea was present. Vomiting was observed in 52% of all the cases, while abdominal pain, cramps, cyanosis and aphonia were also predominant symptoms.

Carriers and carrier rates

The cholera carrier rate in each of the vaccine groups is indicated in Table 26. A total of 111 carriers were found among 1849 contacts, a number deemed insufficient for conclusions. While the carrier rate in the cholera vaccine group was lower than that in the control group, the difference was not significant.

The variation of the number of carriers in each vaccine group with the time interval from vaccination

TABLE 18

GEOGRAPHIC DISTRIBUTION OF CHOLERA CASES, 6 JUNE 1964 TO 5 DECEMBER 1965

Town or city	Population (×1 000)	1964							1965												Total No. of cases	Rate per 1 000
		June	July	Aug.	Sept.	Oct.	Nov.	Dec.	Jan.	Feb.	March	April	May	June	July	Aug.	Sept.	Oct.	Nov.	Dec.		
San Carlos City	139	—	—	3	13	16	76	24	13	2	1	—	—	—	—	—	—	—	—	—	148	1.06
Calatrava	76	—	—	—	—	6	9	12	11	1	—	—	—	—	—	—	—	—	—	—	39	0.51
Toboso	42	—	—	1	—	14	19	11	8	2	2	—	—	—	—	—	—	—	—	—	57	1.36
Escalante	69	—	1	1	—	28	41	11	6	—	1	—	—	—	—	—	—	—	—	—	89	1.29
Sagay	82	—	3	6	5	36	28	68	52	30	—	—	—	—	—	—	—	—	—	—	228	2.78
Cadiz	102	—	6	5	3	1	7	26	30	7	—	—	—	—	—	—	—	—	3	—	88	0.86
Manapla	54	—	—	—	—	2	4	9	10	3	—	3	—	—	—	—	—	—	4	—	35	0.65
Victorias	40	—	—	—	2	2	6	18	4	3	1	—	—	—	—	—	—	2	16	—	54	1.35
Saravia	37	—	2	1	3	7	12	19	5	—	—	—	—	—	—	—	—	2	2	—	53	1.43
Silay City	70	—	—	8	9	9	16	30	6	—	—	—	—	—	—	—	—	7	17	—	102	1.46
Talisay	54	—	3	15	7	4	21	40	14	1	1	—	—	—	—	—	—	11	26	—	143	2.65
Bacolod City	138	10	37	46	34	25	42	73	18	4	—	—	—	—	1	—	—	3	29	—	322	2.33
Murcia	27	—	1	6	2	—	12	17	8	1	—	—	—	1	—	—	—	1	1	—	50	1.85
Bago	68	1	6	15	25	14	13	9	3	—	—	—	—	—	—	—	1	—	1	—	88	1.29
Pulupandan	18	—	4	6	8	8	3	2	1	—	—	—	—	—	—	—	—	—	—	—	32	1.78
Valladolid	17	—	22	8	11	2	—	7	—	—	—	—	—	—	—	—	—	—	1	—	51	3.00
San Enrique	12	—	2	3	4	6	1	—	2	—	—	—	—	—	—	—	—	—	—	—	18	1.50
Pontevedra	26	—	2	2	2	1	2	2	—	1	—	—	—	—	—	—	—	1	—	—	13	0.50
Hinigaran	43	4	31	6	14	14	5	2	—	—	—	—	—	—	—	—	—	—	—	—	76	1.77
Binalbagan	36	—	9	6	8	1	2	4	—	—	—	—	—	—	—	—	—	—	—	—	30	0.83
Isabela	34	1	2	4	9	6	3	—	4	—	—	—	—	—	—	—	—	—	—	—	29	0.84
Himamaylan	49	1	15	9	18	11	2	1	2	—	—	—	—	1	—	—	—	—	—	—	60	1.22
Kabankalan	68	—	21	7	6	10	1	6	7	—	—	—	—	—	—	2	—	—	—	—	60	0.88
Ilog	28	—	5	4	3	6	2	1	1	—	—	—	—	—	—	—	—	—	—	—	22	0.79
Total	1 329	17	172	162	186	229	327	392	205	55	6	3	—	2	1	2	1	27	100	—	1 887	1.42

TABLE 19

CONFIRMED CHOLERA CASES, BY PLACE OF CONFINEMENT, 6 JUNE 1964 TO 5 DECEMBER 1965

Vaccine group	Place of confinement				Total
	Hospital		Home		
	No. of cases	Percentage	No. of cases	Percentage	
Classical cholera	90	68.7	41	31.3	131
El Tor	82	72.6	31	27.4	113
Oil-adjuvant	61	72.6	23	27.4	84
Control	133	77.8	38	22.2	171
Total	366	73.3	133	26.7	499

TABLE 20

DEATHS AMONG VACCINATED PERSONS, 6 JUNE 1964 TO 5 DECEMBER 1965

Vaccine group	No. vaccinated	No. of deaths	Death rate per 100 000
Classical cholera	145 500	13	8.9
El Tor	148 100	10	6.8
Oil-adjuvant	143 600	3	2.1
Control	146 800	13	8.9
Total	584 000	39	6.7

TABLE 21

DEATHS IN EACH VACCINE GROUP, BY TIME INTERVAL FROM VACCINATION TO DEATH, 6 JUNE 1964 TO 5 DECEMBER 1965

Days from vaccination to death	No. of deaths in vaccine group				Total
	Classical cholera	El Tor	Oil-adjuvant	Control	
0-180	9	5	2	11	27
181-360	2	3	0	2	7
361-540	2	2	1	0	5
Total	13	10	3	13	39

TABLE 22

CASE-FATALITY RATE, BY VACCINE GROUP, 6 JUNE 1964 TO 5 DECEMBER 1965

Vaccine group	No. of cases	No. of deaths	Case-fatality rate (%)
Classical cholera	131	13	9.9
El Tor	113	10	8.8
Oil-adjuvant	84	3	3.6
Control	171	13	7.6
Total	499	39	7.8

TABLE 23

AGE DISTRIBUTION OF DEATHS AND FATALITY RATES IN THE 4 VACCINE GROUPS, 6 JUNE 1964 TO 5 DECEMBER 1965

Age-group (years)	Classical cholera vaccine group			El Tor vaccine group			Oil-adjuvant vaccine group			Control vaccine group		
	No. of cases confirmed	No. of deaths	Fatality rate	No. of cases confirmed	No. of deaths	Fatality rate	No. of cases confirmed	No. of deaths	Fatality rate	No. of cases confirmed	No. of deaths	Fatality rate
0-9	72	11	15.3	61	7	11.5	45	4	8.9	76	11	14.5
⩾10	59	2	3.4	52	3	5.8	39	1	2.6	95	2	2.1

TABLE 24

DURATION OF DIARRHOEA IN CONFIRMED CHOLERA CASES IN THE 4 VACCINE GROUPS (DEATHS EXCLUDED), 6 JUNE 1964 TO 5 DECEMBER 1965

Duration of diarrhoea (days)	No. of cases in vaccine group				
	Classical cholera	El Tor	Oil-adjuvant	Control	Total
1	23	22	21	26	92
2	39	20	22	31	112
3	19	20	17	41	97
4	13	15	10	27	65
5	13	4	6	13	36
6	4	2	3	8	17
7	3	3	0	5	11
8	1	5	1	2	9
9	0	1	0	2	3
10	1	0	1	1	3
>10	2	0	0	2	4
Total	118	92	81	158	449

TABLE 25

CLINICAL SYMPTOMS AMONG CONFIRMED CHOLERA CASES IN THE 4 VACCINE GROUPS, 6 JUNE 1964 TO 5 DECEMBER 1965

Symptom	No. of cases in vaccine group			
	Classical cholera	El Tor	Oil-adjuvant	Control
Diarrhoea	131	113	84	171
Vomiting	87	76	52	15
Abdominal pain	29	18	28	51
Cramps	38	33	28	64
Cyanosis	38	36	26	48
Aphonia	29	23	21	57
Anuria	4	3	3	4
Tympanism	0	1	0	1
Fever	0	0	0	0
Total	131	113	84	171

to detection shows approximately the same trend as the incidence (Table 21).

Table 27 shows the distribution of cholera carriers by month of detection; this again follows the pattern of endemicity. During the low-incidence periods in March and June, no cholera carriers were detected.

Table 28 gives the carrier rates among household contacts of both hospitalized and home cases in the 4 vaccine groups. About 20% of all the carriers were found among the contacts of home cases. The carrier rate in home cases is apparently about the same as in hospitalized cases.

REACTIONS TO VACCINATION

Reactions to inoculation were observed in 1000 individuals selected at random. About 250 persons in each vaccine group were observed. Examinations were made on the first, second and third days after vaccination. The reactions manifested were erythema, swelling, pain, induration, fever and a feeling of weakness. The vaccinees were equally affected by the different vaccines, except that erythema and

TABLE 26

CONTACT CARRIERS IN EACH VACCINE GROUP, BY TIME INTERVAL FROM VACCINATION TO DETECTION, 6 JUNE 1964 TO 5 DECEMBER 1965

Days from vaccination to detection	No. of carriers in vaccine group				Total
	Classical cholera	El Tor	Oil-adjuvant	Control	
0-30	1	2	1	5	9
31-60	2	3	4	3	12
61-90	5	4	4	3	16
91-120	2	4	3	5	14
121-150	2	4	3	10	19
151-180	6	4	3	4	17
181-210	3	3	—	6	12
211-240	—	3	2	2	7
241-270	—	—	1	—	1
271-300	—	1	—	—	1
301-330	—	—	—	—	—
331-360	—	—	—	—	—
361-390	—	—	—	—	—
391-420	—	—	—	—	—
421-450	—	—	—	—	—
451-480	—	—	—	—	—
481-510	—	—	—	—	—
511-540	2	1	—	—	3
Total	23	29	21	38	111

TABLE 27

DISTRIBUTION OF CHOLERA CARRIERS BY MONTH OF DETECTION, 6 JUNE 1964 TO 5 DECEMBER 1965

Month of detection	Classical cholera vaccine group	El Tor vaccine group	Oil-adjuvant vaccine group	Control vaccine group	Total
June 1964	—	—	—	—	—
July	3	4	4	8	19
August	2	2	3	1	8
September	4	5	3	3	15
October	1	6	3	7	17
November	6	4	4	9	23
December	5	3	1	8	17
January 1965	—	2	2	—	4
February	—	1	1	2	4
March	—	1	—	—	1
April	—	—	—	—	—
May	—	—	—	—	—
June	—	—	—	—	—
July	—	—	—	—	—
August	—	—	—	—	—
September	—	—	—	—	—
October	—	—	—	—	—
November	2	1	—	—	3
December	—	—	—	—	—
Total	23	29	21	38	111

TABLE 28

CARRIER RATES AMONG HOUSEHOLD CONTACTS OF HOSPITALIZED AND HOME CASES, 6 JUNE 1964 TO 5 DECEMBER 1965

Vaccine group	Hospitalized cases			Home cases			All cases		
	No. of carriers	Total No. of contacts	Rate (%)	No. of carriers	Total No. of contacts	Rate (%)	No. of carriers	Total No. of contacts	Rate (%)
Classical cholera	19	383	5.0	4	105	3.8	23	488	4.7
El Tor	22	380	5.8	7	79	8.9	29	459	6.3
Oil-adjuvant	15	363	4.1	6	81	7.4	21	444	4.7
Control	32	372	8.6	6	86	7.0	38	458	8.3
Total	88	1 498	5.9	23	351	6.6	111	1 849	6.0

TABLE 29

VACCINATION REACTIONS, BY AGE, 6 JUNE 1964 TO 5 DECEMBER 1965

Age-group (years)	No. of reactions	Reactions in vaccine group							
		Classical cholera		El Tor		Oil-adjuvant		Control	
		No.	%	No.	%	No.	%	No.	%
<1	18	2	11.1	0	—	15	83.3	1	5.6
1	75	0	—	0	—	74	98.7	1	1.3
2	71	1	1.4	2	2.8	67	94.4	1	1.4
3	61	2	3.3	1	1.6	57	93.4	1	1.6
4	71	0	—	2	2.8	67	94.4	2	2.8
5-9	470	4	0.9	9	1.9	452	96.2	5	1.1
10-14	652	3	0.5	9	1.4	630	96.6	10	1.5
15-19	278	3	1.1	3	1.1	266	95.7	6	2.2
20-24	245	2	0.8	1	0.4	240	98.0	2	0.8
25-29	248	1	0.4	3	1.2	242	97.6	2	0.8
30-34	188	0	—	0	—	187	99.5	1	0.5
35-39	141	0	—	1	0.7	138	97.9	2	1.4
40-44	117	0	—	1	0.9	116	99.1	0	—
45-49	104	1	1.0	1	1.0	102	98.1	0	—
50-54	90	0	—	0	—	90	100.0	0	—
55-59	36	0	—	1	2.8	34	94.4	1	2.8
60-64	42	0	—	1	2.4	41	97.6	0	—
65-69	7	0	—	0	—	7	100.0	0	—
70-74	14	0	—	1	7.1	13	92.9	0	—
⩾75	6	0	—	0	—	6	100.0	0	—
Total	2 934	19	0.6	36	1.2	2 844	96.9	35	1.2

induration were more pronounced in those given the oil-adjuvant vaccine.

After the first week, it was noted that many of the vaccinees reported to health centres for treatment of abscesses or ulcers due to vaccination. A rapid survey of the study area revealed that many vaccinees developed hard masses ranging from 1 cm to 8 cm in diameter at the site of vaccination. It was further observed that during the succeeding months some of these hard masses showed signs of fluctuation and, if not treated, later developed into ulcers. Fluctuating masses were aspirated by means of a syringe. Smears and cultures of abscesses were found to be bacteriologically negative. The hard masses often subsided after aspiration or disappeared completely, but sometimes, even after repeated aspirations, they developed into ulcers that healed slowly. Vaccinees who exhibited persistent hard masses over a period of time were requested to report to the headquarters in Bacolod for excision of the mass. To date, 2934 vaccinees have shown severe reactions ranging from a hard mass to an abscess or ulcer.

The severe vaccination reactions are analysed in Tables 29, 30 and 31. The reactions included abscesses, ulcers, ulcers with proud flesh, and hard masses. These 2934 severe reactions were evenly distributed over the study area. Calatrava, Escalante, Ilog, Kabankalan and San Carlos City had, however, comparatively low rates of reaction, for which there was no apparent explanation.

TABLE 30

NUMBER OF VACCINATION REACTIONS, BY VACCINE GROUP AND TIME INTERVAL FROM VACCINATION TO FIRST CLINIC VISIT, 6 JUNE 1964 TO 5 DECEMBER 1965

Months from vaccination to first clinic visit	No. of reactions	Reactions in vaccine group			
		Classical cholera	El Tor	Oil-adjuvant	Control
<1	13	0	0	11	2
1	284	1	4	277	2
2	569	4	10	547	8
3	586	7	8	564	7
4	468	3	8	452	5
5	234	0	2	229	2
6	139	1	1	136	1
7	126	1	2	122	1
8	124	0	1	120	3
9	102	1	0	100	1
10	42	0	0	42	0
11	67	0	0	66	1
12	51	0	0	51	0
13	47	0	0	46	1
14	31	1	0	29	1
15	36	0	0	36	0
16	11	0	0	11	0
17	5	0	0	5	0
Total	2 935	19	36	2 844	35

From Table 29 it may be seen that a very high percentage (96.9%) of the total number of severe reactions were caused by the oil-adjuvant vaccine. The cholera vaccine accounted for only 0.6% of the total number of severe reactions, the El Tor vaccine for 1.2% and the control vaccine for 1.2%. Of the 143 600 persons given oil-adjuvant vaccine, 2844 developed severe reactions—a ratio of 1:51.

DISCUSSION

It is held by some that after vaccination with cholera vaccine, human immunity to cholera may last 6 months or more. Others feel that the vaccine protects human beings for less than 5 months (Pollitzer, 1959). However, such opinions are not based upon the results of controlled field trials (Cvjetanovic, 1965).

The present controlled field trial indicated that vaccination with one dose of either of 2 routine types of fluid cholera vaccines, classical or El Tor, produced short periods of immunity. The maximum efficacy of either classical or El Tor vaccine lasted 2 months after vaccination and then decreased gradually. Little, if any, immunity was observed beyond 3 or 5 months after vaccination.

On the other hand, the classical cholera oil-adjuvant vaccine offered approximately 50%–60% protection for the first 6 months, after which the percentage protection decreased gradually. Accordingly, the oil-adjuvant vaccine gave the highest and longest immunity among the vaccinees tested in the present trial. However, because the oil-adjuvant vaccine produced severe reactions in 1.9% of 143 600 vaccinees, it cannot be recommended for practical use.

In fact, the duration of immunity produced by any of the 3 vaccines tested was not sufficient to warrant its use as a mass vaccine. The Calcutta trial (Sinha et al., 1967), in which 4 cholera vaccines were tested, produced the same short-term immunity in humans, indicating that many cholera vaccines available at present prepared according to the WHO minimum requirements (WHO Study Group on Requirements for . . . Cholera Vaccine, 1959), have limited value in cholera control.[1]

Nevertheless, the present trial has provided some important information.

It has been suggested that classical cholera vaccine is able to protect human beings against cholera El Tor infection. Although El Tor vaccine protected humans better than classical cholera vaccine against El Tor infection, the difference was not statistically significant. The effect of classical cholera vaccine was prolonged when used with an oil adjuvant.

The above observations seem to indicate the existence of a common protective antigen for human beings against cholera El Tor, not only in El Tor vibrios, but also in *V. cholerae*, although there is no definite evidence as to which specific antigen or antigens have caused such protection.

Secondly, the possibility of prolonging the efficacy of cholera vaccine by adding an adjuvant was suggested. Because of the severe reactions produced

[1] Various vaccines differ to some extent in their efficacy. Some provide a higher degree of protection, such as that used in the field trials in Dacca, which unfortunately was also rather reactogenic (Oseasohn et al., 1965).

TABLE 31

TYPES OF VACCINATION REACTIONS, BY VACCINE GROUP AND TIME INTERVAL FROM VACCINATION TO FIRST CLINIC VISIT, 6 JUNE 1964 TO 5 DECEMBER 1965

Months from vaccination to first clinic visit	Total No. of reactions	Reactions in vaccine group [a]																			
		Abscess				Ulcer				Ulcer with proud flesh				Tumour				Hard mass			
		Cl	ET	OA	Co	Cl	ET	OA	Co	Cl	ET	OA	Co	Cl	ET	OA	Co	Cl	ET	OA	Co
<1	13	0	0	5	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	6	1
1	284	1	1	91	2	0	1	30	0	0	0	5	0	0	0	0	0	0	2	151	0
2	569	1	3	161	2	0	1	67	0	0	0	21	0	0	0	1	1	3	6	297	5
3	586	2	1	211	3	1	2	114	0	1	0	29	0	0	0	4	0	3	5	206	4
4	468	0	0	152	1	0	0	113	1	1	1	32	0	0	1	1	0	2	6	154	3
5	234	0	1	103	1	0	0	52	0	0	0	21	1	0	0	2	0	0	1	51	0
6	139	0	0	63	0	0	0	26	0	0	0	22	0	0	0	5	0	1	1	20	1
7	126	0	0	47	0	0	0	29	0	0	0	10	0	0	0	2	0	1	2	34	1
8	124	0	0	52	0	0	0	18	0	0	0	23	0	0	0	4	0	0	1	23	3
9	102	0	0	40	0	1	0	7	0	0	0	7	0	0	0	0	0	0	0	46	1
10	42	0	0	13	0	0	0	9	0	0	0	2	0	0	0	0	0	0	0	18	0
11	67	0	0	31	1	0	0	10	0	0	0	7	0	0	0	0	0	0	0	18	0
12	51	0	0	17	0	0	0	4	0	0	0	6	0	0	0	0	0	0	0	24	0
13	47	0	0	16	0	0	0	5	1	0	0	5	0	0	0	1	0	0	0	19	0
14	31	0	0	6	1	0	0	3	0	0	0	4	0	0	0	0	0	1	0	16	0
15	36	0	0	10	0	0	0	5	0	0	0	1	0	0	0	0	0	0	0	20	0
16	11	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	9	0
17	5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	5	0
Total	2 934	4	6	1 018	11	2	4	494	3	2	1	195	1	0	1	20	1	11	24	1 117	19

[a] The vaccine groups are indicated by the abbreviations: Cl = Classical cholera; ET = El Tor; OA = Oil-adjuvant; Co = Control.

by the adjuvant vaccine tested, further studies should be made to find another which can be used without serious reactions in humans.

The relative incidence of the infection was much higher in the younger age-groups. About 28% of all cases occurred in the age-group 1–4 years, while more than 50% of all cases occurred in the age-group 1–9 years. Diminished susceptibility during adolescence is followed by a slight increase during young adulthood. In the older age-groups, a gradual decline in incidence was observed. This is a pattern which is found in practically all endemic cholera areas of the world.

The male-female proportion in confirmed cholera cases shows that both sexes are equally affected by the disease.

It is quite interesting that the 3 vaccines used in this trial gave rather poor protection to the youngest age-group (0.5–9 years), but good protection to the oldest age-group (older than 25 years), although both had higher attack rates than the middle group (10–24 years). The following explanations may be suggested.

(1) The size of the inoculum for the youngest age-group (0.25 ml–0.5 ml for classical and El Tor cholera vaccine, and 0.05 ml–0.1 ml for oil-adjuvant vaccine) may not have been sufficient to give reasonable protection; the doses used for the middle group were 1.0 ml of the former and 0.2 ml of the latter.

(2) The immune response of the youngest group may have been inferior to that of the older groups.

In order to answer such questions, further study of the inoculum size and dose schedule will be required.

Out of 499 cases, 39 terminated in death. The death rates in the classical and the vibrio El Tor cholera vaccine groups was almost the same as that in the control group. However, mortality in the oil-adjuvant group was so low as to be remarkable in comparison with other vaccine groups. It is tempting to draw conclusions on the strength of these data but this must be deferred until more data become available, to increase the statistical significance of the results.

CONCLUSIONS

The classical and El Tor cholera vaccines which are safe to use confer low protection. The duration of the protective effect of the classical cholera vaccine is only 2 months while that of the vibrio El Tor is 4 months. An oil-adjuvant vaccine was definitely protective for a period of 6 months but its protective value beyond that period could not be assessed properly because of the scarcity of cases. However, in view of the severe reactions which it caused, this vaccine cannot be recommended at this time. Further laboratory and field investigations are necessary to improve and assess the value of different types of cholera vaccines. This is particularly important if the control of cholera is to be effected primarily through vaccination programmes.

The relative incidence of cholera is much higher in the younger age-groups but is equal for males and females.

Accurate assessment of the clinical course of the disease was not possible during this study.

The cholera carrier rate appears to be decreased by vaccination, though not significantly.

The carrier rate among the contacts of the home cases is apparently comparable to those of hospitalized cases.

ACKNOWLEDGEMENTS

The authors are deeply grateful to Governor Gomez, to the late Governor Gaduslao, Mayor Guanzon of Bacolod City, and to the people of Negros Occidental Province for the support and co-operation extended to the workers in the field.

Special acknowledgement is hereby made of the important contribution made by Dr Keizo Nobechi of Japan to the planning and evaluation of the study.

Thanks are due to Dr Hideo Fukumi, Dr Hiroshi Ogonuki, Dr Shin-ichi Matsuda, Dr Ei-ichi Nakano, Dr Kan Matsushita, Dr Masami Nagao, Dr Yoshio Chikasato, and Dr Koyuro Sakaguchi from Japan; to Dr V. Mauricio, Dr F. Jayme, Dr J. Suva, Dr G. Justiniano, Dr R. Navarro, Miss D. Gaetos, Mrs E. G. Maniago, Miss L. Policarpio, Mr G. Penaflor and the field staff, from the Philippines for their valuable assistance in the various facets of this project.

RÉSUMÉ

Depuis l'apparition du premier vaccin anticholérique proposé par Ferran en 1885, aucun essai pratique strictement contrôlé de ce genre de préparations n'avait été fait. Les études collectives actuellement en cours sur l'efficacité des vaccins anticholériques ont commencé en 1964, à la suite d'un accord entre le Gouvernement des Philippines, le Gouvernement du Japon et l'Organisation mondiale de la Santé, sur un échantillon de 584 026 sujets constitué à partir d'une population de 1 700 000 habitants de la province du Negros occidental (Philippines).

Les objectifs en étaient les suivants: évaluer le pouvoir immunisant des vaccins à l'égard des manifestations cliniques du choléra El Tor et des infections asymptomatiques par les vibrions El Tor; apprécier le degré et la durée de la protection conférée par chaque type de vaccin; et étudier pour chacun d'eux les réactions postvaccinales. Les essais ont été organisés selon des principes très stricts. L'efficacité de chaque vaccin a été évaluée par comparaison du taux de morbidité et du taux de porteurs relevés pour les biotypes El Tor chez des sujets vaccinés au moyen soit d'un vaccin anticholérique, soit d'un vaccin sans aucun lien antigénique avec les vibrions du choléra classique ou ceux du choléra El Tor. Deux des vaccins anticholériques utilisés étaient des vaccins liquides en milieu aqueux, l'un préparé à partir de souches de *Vibrio cholerae* et l'autre à partir de cultures lyophilisées de vibrions El Tor; un vaccin anticholérique en excipient huileux a été également mis à l'essai, et un vaccin antityphoïdique monovalent a servi de placebo. La comparabilité des résultats a été assurée par une répartition rigoureusement aléatoire des vaccins.

Ces essais pratiques ont montré que le vaccin anticholérique classique et le vaccin El Tor, qui sont inoffensifs, ne confèrent qu'une faible protection. La durée de l'immunité est de deux mois pour le vaccin anticholérique classique et de six mois pour le vaccin El Tor. Le vaccin huileux donnait une protection certaine pendant six mois; son pouvoir immunisant au-delà de cette période n'a pu être évalué du fait de la rareté des cas. En raison, toutefois, de la gravité des réactions qu'il provoquait, on ne saurait en recommander l'emploi généralisé pour le moment. Il faudra de nouvelles études en laboratoire et sur le terrain pour juger de la valeur relative des différents types de vaccins anticholériques et pour les améliorer. Le taux d'incidence du choléra El Tor est apparu beaucoup plus élevé dans les groupes d'âges les plus jeunes, mais il était similaire pour les deux sexes. Il n'a pas été possible, au cours de cette étude, d'apprécier avec exactitude l'évolution clinique de la maladie. Il semble que la vaccination ait entraîné une diminution du pourcentage de porteurs, quoique assez peu marquée. Le pourcentage de porteurs parmi les contacts des malades soignés à domicile était comparable à celui observé parmi les contacts des cas hospitalisés; l'un et l'autre étaient plus élevés que le pourcentage constaté dans l'ensemble de la population.

REFERENCES

Cvjetanovic, B. (1965) *Earlier field studies of the effectiveness of cholera vaccines.* In: *Proceedings of the Cholera Research Symposium, Honolulu,* Washington, D.C., US Government Printing Office, p. 355

Ferran, J. (1885) *Siglo méd.*, **32**, 480

Joseph, P. R., Tamayo, J. F., Mosely, W. H., Alvero, M. G., Dizon, J. J. & Henderson, D. A. (1965) *Bull. Wld Hlth Org.*, **33**, 637

Nobechi, K. (1965) *Joint Philippines–Japan–WHO controlled field trials of cholera vaccines (interim report of Dec. 5, 1964).* In: *Proceedings of the Cholera Research Symposium, Honolulu,* Washington, D.C., US Government Printing Office, p. 366

Ogonuki, H., Hashizume, S. & Takashashi, B. (1967) *Bull. Wld Hlth Org.*, **37**, 729

Oseasohn, R. O., Benenson, A. S. & Fahimuddin, M. (1965) *Cholera vaccine field trial in rural East Pakistan (first year of observation).* In: *Proceedings of the Cholera Research Symposium, Honolulu,* Washington, D.C., US Government Printing Office, p. 362

Pesigan, T. P., Sumpaico, J. & Sychangco, G. (1967) *Bull. Wld Hlth Org.*, **37**, 816

Philippines Cholera Committee (1965) *Bull. Wld Hlth Org.*, **32**, 603

Pollitzer, R. (1959) *Cholera,* Geneva (*World Health Organization: Monograph Series,* No. 43)

Sinha, R., Deb, B. C., De, S. P., Abou-Gareeb, A. H. & Shrivastava, D. L. (1967) *Bull. Wld Hlth Org.*, **37**, 89

WHO Study Group on Requirements for ... Cholera Vaccine (1959) *Wld Hlth Org. techn. Rep. Ser.*, **179**