



WORLD HEALTH ORGANIZATION
ORGANISATION MONDIALE DE LA SANTÉ

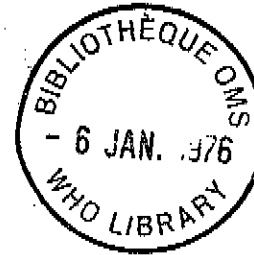
BST/POLIO/75.1

ENGLISH ONLY

RESTRICTED

WHO CONSULTATIVE GROUP ON ORAL
POLIOMYELITIS VACCINE (SABIN STRAINS)

Geneva, 3-5 November 1975



INDEXED

REPORT

The Director-General welcomed the members of the Consultative Group and the consultants to the meeting. He thanked them for the great deal of time that they give to the Organization throughout the year in ensuring that poliomyelitis vaccine continues to be produced correctly throughout the world.

Professor J.L. Melnick was elected Chairman, Dr S.G. Drozdov, Vice-Chairman, and Drs I. Dömök and J. Furesz were elected joint Rapporteurs. A list of the members of the Consultative Group in attendance, together with the consultants and observers is given in Annex I and the Agenda is attached in Annex 2.

1. WHO inquiry into acute persisting spinal paralysis

Although eleven countries have taken part in the study, one country (Denmark) was omitted from the analysis because of the special vaccination schedule used. Five countries gave the vaccine in fixed short-period campaigns, and five gave the vaccine throughout the year (Table 1).

A total of 362 cases of acute spinal paralysis persisting for more than six weeks, 231 in the short-period vaccination countries and 131 in the throughout-the-year vaccination countries, were reported to WHO over the period 1970-1974 inclusive. Out of these cases, 206 were associated in time with vaccination.¹ From an analysis of the findings it appears that in the five countries with short-period vaccination campaigns, infants who receive the vaccine, together with the unvaccinated children in contact with them, form the majority of the cases reported. Of the 166 cases associated in time with polio vaccine, 155 occurred in the age group below five years. In contrast, in the other countries, there were 40 such cases and 22 occurred in the age group 15 years and over and most of these cases were in adults exposed to their own vaccinated children.

In both groups of countries the recipient cases were associated mainly with type 3 (Table 2). Fifteen of 25 type 3 cases were in countries with short-period vaccination, and 4 of 7 type 3 cases were in the other group of countries. In contrast, the contact cases in both groups of countries were associated more frequently with the type 2 virus, 52/76 in both the contact and possible contact cases in the short-period countries, and 12/21 for contact cases in the throughout-the-year countries.

In the group of countries with short-period vaccination campaigns there is no preponderance of poliovirus type in the 'no known contact' cases, whereas in the other countries most of the cases were due to type 1 poliovirus (Table 2) and the majority of these occurred in two outbreaks in poorly-vaccinated populations in one country.

¹It should be noted that temporal association of a case with vaccination does not necessarily mean that the case was caused by the vaccine virus.

The issue of this document does not constitute formal publication. It should not be reviewed, abstracted or quoted without the agreement of the World Health Organization. Authors alone are responsible for views expressed in signed articles.

Ce document ne constitue pas une publication. Il ne doit faire l'objet d'aucun compte rendu ou résumé ni d'aucune citation sans l'autorisation de l'Organisation Mondiale de la Santé. Les opinions exprimées dans les articles signés n'engagent que leurs auteurs.

The comparison of the experience of the collaborating countries with short-period vaccination programmes annually and those where vaccination proceeds throughout the year has confirmed previous observations that in the short-period countries cases are concentrated in the period immediately following the use of the vaccine, but in the other countries the cases occur throughout the year.

When the study was initiated in 1970, cases following the administration of type 3 vaccine were the most frequently reported. Since 1972, however, cases associated with type 2 are the most frequently encountered, particularly in contacts of vaccinated children.

It was recognized that the notification of cases and the intensity of investigations differ from one country to another. Nevertheless, the results over the five years of this study have justified the very detailed inquiries made in the collaborating countries.

The consultative group recommended that:

- 1.1 The study should be continued and expanded by inviting other countries to take part.
- 1.2 Cases should be investigated in more detail, particularly concerning
 - a) the cellular content of spinal fluid during the first 10 days after the onset of paralysis, and
 - b) the immuno-deficiency states.
- 1.3 Serological surveys should be carried out to determine the immunity profile of the population.
- 1.4 The report be published along an outline prepared by the group (see Annex 3).
2. Collaborative studies on markers of poliovirus strains

The investigations on 38 selected poliovirus strains originating from 28 patients observed in 1970 and 1971 in five countries were continued. Although all the collaborators were not able to carry out all the tests recommended at the last consultation in 1974, the results that were available were sufficiently satisfactory to make an analysis of the strains in respect of their antigenic and elution marker characters from which the following conclusions were drawn:

- 2.1 Four type 1 strains originating from "recipient", "contact" and "no known contact" cases which occurred in 1970 in Poland were different from the vaccine LSc 2ab strain in respect of their antigenic and elution character. This was not surprising because the Chat strain was included as the type 1 component of the live polio vaccine used in Poland in 1970, but it was an interesting observation.
- 2.2 Eleven of 14 type 2, and 17 of 19 type 3 poliovirus isolates - all originating from recipient or contact cases - proved to be vaccine-like both in intratypic serodifferentiation and in elution marker tests. With certain strains, however, some discrepancies in the results obtained from different laboratories were observed.
- 2.3 Three of the type 2, two of the type 3 and one of the type 1 isolates remained uncharacterized in respect of their antigenic characteristics because there were significant differences in the results obtained from the collaborating laboratories. Some of these isolates were actually mixtures of two different types of poliovirus which explains, at least in part, the difficulties in characterizing them.

- 2.4 As the study progressed it appeared that the original technique described for the aluminium $Al(OH)_3$ elution marker gave incorrect results especially for vaccine derived strains because it was shown that, in addition to the viruses there remained cell constituents and these were also labelled with ^{32}P causing a shift in elution coefficient 50% (EC50) values. As a result of these observations the modified technique that was then introduced proved to be suitable to characterize all three types of poliovirus strains.
- 2.5 Model experiments carried out with vaccine and wild strains demonstrated that a 10-minute period of incubation was satisfactory to obtain reproducible results in the McBride tests.

The group recommended that the IS and elution marker test studies should be continued as follows:

- 2.6 Some strains originating from countries from which no strains or only a few strains were investigated so far (England and Wales, Scotland, Japan, the USA, Canada) should be included.
- 2.7 The validity of the modified $Al(OH)_3$ elution marker test should be checked by testing a number of strains isolated in countries prior to the introduction of the use of live vaccine.
- 2.8 The McBride test should be included using only a single 10-minute period of incubation of serum virus mixtures.
- 2.9 The offer of Dr Cohen of R.I.V., Bilthoven, to supply the collaborators with adsorbed strain specific sera was accepted, but it was felt that before distribution of the sera to the collaborators it was necessary to test them against wild strains with different origins. It was agreed to supply the Bilthoven laboratory with control strains of all three types of poliovirus. If the tests, using a simple neutralization technique, indicated that the sera were useful for the differentiation of vaccine strains from wild strains, then the sera would be distributed to the collaborators. The collaborating laboratories will then be asked to test the strains using these sera by the McBride or classical neutralization technique.
3. Present situation in several countries concerning the production of vaccine
- 3.1 Egypt (Agouza Institute). It was found that this area was not yet fully equipped or staffed to undertake the production of poliomyelitis vaccine and, therefore, it was suggested that the staff should gain experience by testing imported vaccine. Steps were being taken to do this. In order to help the laboratory to prepare final vaccine from imported bulk, the group agreed that there should be precise instructions written for the diluting, blending and stabilizing of vaccine. The Secretariat undertook to circulate a draft of such instructions.
- 3.2 Iran (Razi Institute). These laboratories were considered to be satisfactory in all respects. The technical achievements were of a high standard and although short of staff they had produced their first six consecutive lots (2 of each virus type) which had been tested by external control laboratories. It was verbally reported that all tests had been shown to be satisfactory and as soon as the written reports were received the Secretariat would write to the Institute informing them of the results. The Institute intended to continue subjecting their future batches to external control until such time as the Ministry of Health in Iran established a control laboratory.

- 3.3 India. The situation in India was still unsettled.
- 3.3.1 The Haffkine Institute, Bombay, had declared its intention to start producing vaccine but the area was found to be unsuitable and unacceptable. Plans of a new building have been submitted to WHO and the area is to be revisited in November 1975, at which time discussions with the Ministry and with the Institute staff will take place.
- 3.3.2 The Pasteur Institute, Coonoor. Although vaccine preparation had been in progress for over 10 years, very little had been produced. Some batches, stored in the bulk form, had lost virus titre because of frequent interruptions of supplies of electricity. It was felt that such a long time had elapsed between the early production and the recent preparation of vaccine that the laboratory must re-establish a record for consistency of production. The director of the laboratory has been informed of this decision in writing.
- 3.3.3 The National Institute for Communicable Diseases, New Delhi, carried out neurovirulence tests as the external laboratory control function for vaccine produced in India. The senior scientist and his staff were well trained and had offered their facilities to the Group for the testing of vaccines from other countries. The Group agreed that this offer should be explored.
- 3.4 Mexico (Institute of Virology). Many improvements in the facilities had been made in order to bring them up to modern standards. The staff were well trained and knowledgeable of the WHO Requirements for vaccine production and testing. The first six consecutive lots of vaccine (all type 1) had passed all tests both by the staff and by an external control laboratory, and they had now been approved by the assessor to PAHO. As a result of this, the WHO type 3 and the former type 2 seed viruses had been sent to them.
- 3.5 Poland (Serum and Vaccine Manufacturing Establishment, Lublin). The staff of the production and control laboratory had now received training abroad and they were in a position to produce vaccine. The laboratories had undergone modifications and were considered suitable for vaccine production. The WHO type 3 and the former type 2 seed viruses had been supplied to the laboratory and the first lots of vaccine were currently being produced and tested.
- 3.6 Romania (Cantacuzino Institute, Bucharest) had moved their production facilities to an entirely new building. This area was satisfactory and had been used to produce vaccine from the WHO type 3 seed virus which had been used as the type 3 component of trivalent vaccine for vaccination throughout the country in 1975; no untoward reactions had occurred.
- 3.7 USSR (Institute of Poliomyelitis and Virus Encephalitides, Moscow) had built a new production facility in 1973 which was satisfactory. The staff were well trained and had much experience. There were some divergencies from the WHO Requirements in the testing of the vaccine but it was agreed that in future these would be changed to those required by WHO. It had been agreed that some batches of each type of vaccine and the seeds would be made available for testing by an independent laboratory.

4. Distribution of WHO type 2 and type 3 seed viruses

Type 3. Since 1970, 105 ampoules of the WHO type 3 seed virus (Glaxo seed pool 6, SO+1) have been distributed to five manufacturers (Mexico, Burroughs Wellcome, Warsaw, Bucharest and Pfizer). In Romania a large batch of vaccine (L.U. 3 - 1/74 at SO+2 passage) was made in 1974. In 1975 some of this vaccine was used as the type 3 component of trivalent vaccine for children. There were no untoward reactions and this batch will be used in Romania as the seed for the production of further batches of vaccine.

Type 2. Pfizer batch 129/II, adopted as the WHO type 2 seed, had been available for about one year and only two manufacturers (Warsaw and Mexico) had received it but tests of vaccine made from this material were still in progress. In view of preliminary data reported on further passages of this material showing changes in the rct and neurovirulence characteristics, the Group decided that no vaccine produced from this material should be used until further data were available. Such data would be considered by the Group before making a decision on the future use of this type 2 seed material.

As an interim measure, seed lot No.2 (at the SO+2 passage) obtained from Pfizer will be distributed by WHO for the production of vaccine directly from the material so that vaccine at the SO+3 level will be available. Such a procedure had been used by Pfizer for many years and vaccine at the SO+3 level had been given to millions of subjects.

5. Laboratory tests of a Pfizer and a Russian type 3 vaccine

WHO were interested in the suitability of vaccine prepared from a seed that had been made from the WHO type 3 seed (seed pool 6) but before this Lot No.232-III (Pfizer) was used for mass vaccination in Hungary it was compared in Hungary and the USSR for neurovirulence in monkeys with Lot No. 653 (Moscow).

The conclusion from Hungary and the USSR was that Lot No. 653 was slightly less neurovirulent than Lot No. 232-III. Despite this difference Lot No. 232-III satisfied the WHO Requirements for the neurovirulence of live poliovirus vaccines prepared from the Sabin strains.

When the histological preparations made in Hungary were submitted for examination by two independent pathologists it was concluded that there did not appear to be any difference in the neurovirulence between Lot No. 232 and Lot No. 653.

These findings, coupled with the characteristics of strains re-isolated from infants vaccinated with batch 232-III (see item 6), led to a field trial using this vaccine in children living in a county of Hungary (about 11 000 children) which was started in October 1975. As soon as the results received are evaluated, the vaccination with Lot No. 232-III will be continued in a mass campaign according to a plan agreed with WHO.

6. Small scale trial on Sabin type 3 vaccine, Pfizer Lot No. 232-III

The results of the virologically controlled field trial carried out in four State orphanages in Hungary were reported. Vaccinations with the type 3 vaccine used, prepared in human diploid cells, gave a 70% overall seroconversion and a 75% vaccine virus excretion rate. Thus the vaccine proved to be efficient and clinical observations of children did not reveal any untoward reactions.

Rct marker tests of strains re-isolated from the vaccinees in the early and late stage of excretion indicated that the reversion rate was not higher than those usually observed following the use of type 3 vaccine.

7. Report on the use of vaccine made from the WHO type 3 seed (Glaxo seed pool 6) in Romania

A batch of type 3 vaccine (L.U.3 - 1/74 at the SO+2 level) was prepared by the Cantacuzino Institute using WHO type 3 seed (Glaxo SP6) and was used as the type 3 component of trivalent oral polio vaccine (TOPV) in an extensive field trial conducted in three steps:

- Step 1 : 196 children given the first dose of primary vaccination
- Step 2 : 5018 children given the first dose of primary vaccination
- Step 3 : about 420 000 children given the second dose of primary vaccination
about 80 000 children given one single dose of primary vaccination

All the children were between 6 weeks and 14 months of age. No untoward reactions due to the vaccine were recorded among the recipients. The in vitro marker determinations as well as the intraspinal and intracerebral monkey neurovirulence tests, together with the outcome of this large-scale field test, fully substantiated the safety of L.U.3 - 1/74 and its suitability for use as working seed for the preparation of further batches of type 3 oral polio vaccine (SO+3 passage).

8. Status of collaborative studies of potency testing oral poliovirus vaccines

The attempts of the Bureau of Biologics to develop a standardized technique for assaying the potency of trivalent oral poliovirus vaccine were described. Studies were designed to compare the titrations of a plaque assay and tube test using primary rhesus monkey kidney cells with a microtitre test using HEp-2 (Cincinnati) cells. So far, the microtitre test appeared to give the most reproducible results but the infectivity titres were significantly higher than with the other two procedures using primary monkey kidney cells. The Group considered the need to adjust the potency requirements to compensate for the higher titres, as well as the advisability of having a reference preparation as a control for inter-laboratory results. It was recommended that an international study should be initiated to resolve the problems of potency testing (see Annex 4).

9. Status of the original seed viruses of oral poliovirus vaccine

The original Sabin seed viruses currently stored in bulk at the Bureau of Biologics, USA, have been distributed into ampoules in 0.5 ml aliquots. There were:

- 30 ampoules of type 1 (LSc 2ab, KP₂),
- 10 ampoules of type 2 (P712 Ch2ab-KP₂), and
- 9 ampoules of type 3 (Leon 12a1b, KP₃).

At the time of distribution samples were taken for tests from which it was shown that the sterility tests of each of the seeds were negative.

The original viruses for each virus type were titrated at the Bureau of Biologics, USA. Eagle medium with serum was used as tissue culture fluid and the 7 days readings of virus titre per 0.1 ml were as follows:

	HEp-2 (passage 232) cells	Primary rhesus monkey kidney cells
Type 1	7.4	6.5
Type 2	7.0	6.2
Type 3	6.5	5.5

It was recommended that these materials should not be distributed directly to manufacturers but that they be used for generating stocks of virus at the SO+1 passage which could be made available as WHO primary seeds.

10. Status of HEP-2 (Cincinnati) cells

The current inventory of HEP-2 (Cincinnati) cells at the Bureau of Biologics, USA, is 75 ampoules at passage 142. Stocks of the original passage 135 were lost due to the failure of a liquid nitrogen container. It was recommended that efforts should be made to locate alternative stocks at a lower passage. Studies were in progress to determine whether or not the sensitivity of the cells to infection with poliovirus changed when successively higher passage levels were used for potency testing. Karyologic and isozyme characteristics of the cell line were to be determined and tests for endogenous oncornavirus information would be carried out. The Bureau of Biologics was designated as the repository for the HEP-2 (Cincinnati) cells.

11. Action to be taken to approve laboratories with a long record of production of vaccine

The Secretary of the Consultative Group had approached all the laboratories with whom Professor Sabin had a contractual arrangement to inform them of the transfer of responsibility for supervision for vaccine production to the Director General of WHO.

Some of those manufacturers who had requested to be recognized by the WHO scheme had many years of experience in the production of poliomyelitis vaccine and were approved by their national control authority. Nevertheless the Group agreed that in order to comply with the conditions for approval by WHO, all laboratories should be inspected and a report circulated to the Group for their consideration.

It was agreed also that the records of passage history of seed viruses used by manufacturers should be sent to WHO and the inventory kept up-to-date. It would be useful also to know of the stocks of quantities of seed materials held by each manufacturer.

12. Approval of a new vaccine production technique in Mexico

The development by George Mann in Mexico of a new perfusion culture apparatus for the large scale production of tissue cultures, using human diploid cells, was described.

During a four-year research and development period parameters were established for the optimal conditions for cell production and the propagation of poliovirus of Sabin type 1 (LSc 2ab) strain. It had been found possible not only to double the cell density as compared with stationary bottle cultures but also to increase the yield of virus per cell and per unit area of cell growth by factors of three and five fold.

Three sizes of the production equipment had been made so far giving surface areas of 2.5, 5 and 10 square metres. The apparatus is assembled and sterilized by autoclaving. The largest size was capable of producing about 120 litres of virus suspension of high titre harvested by pooling the harvests at 48, 72 and 96 hours after infection.

Two or more vessels can be coupled in parallel or in series, thus making it feasible to use an in-line vessel of smaller capacity for control testing, the whole assembly operating under identical conditions up to the moment of virus infection. At this time the control vessel is disconnected and sent to the Control laboratory for further maintenance until the time of the third virus harvest when the cells are subjected to a further split in bottles for two further passage levels. Alternatively the control vessel can be used for cell subculture in bottles.

The use of a control vessel with four glass plates of the standard size allows optical examination of six surfaces each having 8×10^7 cells per surface with an area of 2400 sq cms (48×10^7 cells) by using a collar to extend the objective of the microscope. Such an apparatus is under design at this moment.

Although control testing of diploid cell substrates is more extensive than that of monkey kidney produced vaccines, the large volumes of harvest greatly reduces the load on the control laboratories in all respects. The consequent savings on control overheads, purchase of increasingly expensive monkeys from rapidly decreasing supplies, diminution of production laboratory labour, space and service overheads, have been theoretically calculated to effect a saving of more than 90% in production and control costs for poliomyelitis vaccine.

Rate and volume of production as envisaged, if achieved, will release production facilities for sequential production of other viral vaccines for both human and animal use. This will enable other production laboratory space to be released for the modification and provision of the greatly increased cold room storage facilities needed for the production on the huge scale of finished product now in prospect and for the installation of high-speed filling equipment capable of turning out final containers for continental, rather than national, quantities of vaccine.

In liaison with other groups of the Pan American Health Organization parallel parameter determinations for the optimal use of the apparatus have been discussed for the production of animal viral vaccines in Central and South America.

No patent rights or license fees are sought in the use of the new perfusion culture vessel, the principles of which have already been described in the Pan American Health Organization Bulletin (1972), VI (3): 33-36.

As has been the case with the production and control of poliomyelitis vaccine in primary monkey kidney cells, the PAHO consultants involved in the proposed production of viral vaccines in the new apparatus are producing a procedural manual. In consequence of the novel procedures involved, it is anticipated that some modification of WHO requirements for the control of human viral vaccines may be necessary, but these will be discussed at an early stage. A request was made, therefore, that the final manual of procedure, evolved in Mexico, be accepted for perusal by members of the Consultative Group with a view to reaching agreement on such modifications in the requirements as they may consider advisable.

13. Inhibitory substance in human saliva as a factor responsible for low efficacy of live poliovirus vaccine in tropics

As a continuation of studies started in Uganda in 1972 a vaccination trial using type 1 live vaccine was carried out in 1974-75 and the results were presented. The main aim of the trial was to test the significance of the presence of a saliva inhibitor at the time of vaccination by studying the effect on the responses to vaccination. It was found that if the saliva inhibitor was present at the time of vaccination, circulatory and secretory antibody responses, as well as vaccine virus excretion rates, were lower than if the inhibitor was absent. These results were in harmony with those obtained in former studies in Uganda and suggest that the inhibitor found in saliva may be one of the factors responsible for the low efficacy of live vaccine often observed in warm climate countries. The Group felt that the studies should be continued particularly in order to clear up the exact nature, mode of action, action spectrum of the inhibitor and its occurrence in different populations.

14. Testing of neurovirulence of poliovirus vaccines in cynomolgus and rhesus monkeys

The difficulties in interpretation of the criteria currently being used by the Bureau of Biologics, USA, to evaluate the results of neurovirulence tests of poliovirus vaccines were described and a review of the approaches being made to resolve them was presented. The objective was to develop a statistically sound method for quantitating the results of both the intrathalamic and intraspinal tests. The experiences of different laboratories, using either an intrathalamic or intraspinal test or both, were reported and the relative merits of cynomolgus versus rhesus monkeys mentioned. The results of a previous international study¹ of neurovirulence testing methods in monkeys was cited. The effects upon the present consistency requirements of applying the revised criteria, which might reject more lots of vaccine than has been experienced previously were considered. The need to re-evaluate periodically the criteria to assure current day relevance was recognized and WHO proposed holding a meeting for this purpose in 1976. The advantages and disadvantages of developing a neurovirulence testing laboratory as a reference laboratory for WHO was discussed. Although it was agreed that the availability of such a facility would greatly help those countries not having the means to carry out independent testing of vaccines, its existence could not relieve the responsibility that national control authorities had for this safety test. It was suggested that this matter should be explored further to determine if all of the requirements for experienced personnel, animal holding facilities, and the capacity for virological and serological testing, could be satisfied before a laboratory could be considered acceptable.

15. Stability of Sabin's type 2 vaccine strain

Preliminary data concerning the monkey neurovirulence and rct marker character of type 2 Sabin virus (P7₁₂Ch2ab-KP₂) harvests from different sources were reported. Increased activity in the neurovirulence test correlated well with increased ability to replicate at 39°C in rhesus monkey kidney cells.

One of the virus lots was a sample from the Pfizer batch 129 (passage level SO+3) acquired by WHO for distribution as type 2 seed virus. This material had been passaged five times in primary patas monkey kidney cell monolayers at a temperature of 34 ± 0.5°C and the virus seeding level was such that it required three to four days to degenerate the tissues before the virus was harvested. These conditions were chosen deliberately in the belief that if there was any instability in this virus strain then on passage this would be revealed quickly. The third (SO+6) and fifth (SO+8) passages, that is, one and three passages respectively beyond the level at which vaccine would be produced, were selected for study. Both samples were considered to be different from the reference material as shown by their neurovirulence and rct marker characters. Despite the fact that the virus was passaged under conditions not subject to the scrutiny usual in the production of vaccine, the Group thought that these results could not be ignored and that the seed should not be used by producers. The Group approved, as a temporary solution, the purchase from Pfizer of their present type 2 seed (SO+2), prepared in monkey kidney cells, for use as working seed to manufacturers requesting it.

The long term aim was to obtain fresh seeds, of all three types, prepared from the SO material now held at the Bureau of Biologics. Arrangements for this had already been made. When these seeds become available they will be distributed to manufacturers for the preparation of working seed, and thus vaccine would be available at the SO+3 level. The Group suggested that studies of the behaviour of the Sabin strains on cell passage be continued under precisely defined and controlled conditions. Definitive studies should be carried out also on the seeds that are to be made from the SO material.

¹This study was carried out by the International Association of Biological Standardization and took place between Germany, the USA and the UK.

TABLE 1. VACCINE DISTRIBUTION IN PARTICIPATING COUNTRIES

Country	Estimated population mid-1973 (millions)	Number of doses distributed (1970-74)				TOTAL
		MOPVI	MOPV2	MOPV3	BOPV283	
<u>Short-period vaccination countries¹</u>						
Czechoslovakia	15.0	1 699 273	-	-	1 648 747	-
Hungary	10.4	2 555 538	2 659 565	2 446 182	-	-
Japan	107.3	-	-	-	-	19 099 722
Poland	33.0	1 812 360	1 640 989	1 705 857	-	3 580 959
Romania	21.0	1 640 969	-	-	-	10 123 330
TOTAL	186.7	7 708 140	4 300 554	4 152 039	1 648 747	32 804 011
<u>Throughout-the-year vaccination countries²</u>						
Canada	22.5	-	-	-	-	6 206 670
England & Wales	49.0	-	-	-	-	9 630 375
Norway	4.0	-	-	-	-	2 603 030 ³
Scotland	5.2	-	-	-	-	1 849 182
U S A	210.3	-	-	-	-	126 153 339
TOTAL	291.0	-	-	-	-	146 442 596
TOTAL		-	-	-	-	146 442 596

¹ Countries in which vaccination campaigns are carried out at short specific times of the year

² Countries in which vaccination is offered throughout the year

³ Number of doses distributed during the period 1970-73

MOPV = monovalent vaccine
BOPV = bivalent vaccine
TOPV = trivalent vaccine

TABLE 2. REPORTED CASES OF ACUTE PERSISTING SPINAL PARALYSIS

	Short-period vaccination countries					Throughout-the-year vaccination countries				
	Recipient cases	Contact or possible contact cases	Sub-total	No known contact cases	Total	Recipient cases	Contact or possible contact cases	Sub-total	No known contact cases	Total
Total number of cases	48	118	166	65	231	13	27	40	91	131
Diagnosed solely on clinical grounds	11	32	43	31	74	0	2	2	17	19
Cases from which polio-viruses were isolated	37	86	123	34	157	13	25	38	74	112
Cases in which more than one poliovirus was identified	12	10	22	5	27	6	4	10	4	14
Cases in which one poliovirus was identified:										
Type 1	2	5	7	9	16	2	3	5	50	55
Type 2	8	52	60	10	70	1	12	13	12	25
Type 3	15	19	34	10	44	4	6	10	8	18
Total:	25	76	101	29	130	7	21	28	70	98

LIST OF PARTICIPANTS

Members of the Consultative Group^{*}

- Dr I. Archetti, Istituto Superiore di Sanità, Viale Regina Elena 299, Rome, Italy
- Dr I. Dömök, Head, Department of Virology, National Institute of Hygiene, Gyali ut.2-6, H-1966 Budapest IX, Hungary
- Dr S.G. Drozdov, Director, Institute of Poliomyelitis and Virus Encephalitides, P.O. Institute of Poliomyelitis, Moscow Oblast 142782, USSR
- Dr J. Furesz, Director, Bureau of Biologics, Drug Directorate, Tunney's Pasture, Ottawa, Ontario K1A 0L2, Canada
- Professor J. Kostrzewski, Chief, Department of Epidemiology, National Institute of Hygiene, 24 Chocimska Street, Warsaw 36, Poland
- Professor J.L. Melnick, Department of Virology and Epidemiology, Baylor College of Medicine, Houston, Texas 77025, USA
- Professor A.B. Sabin, Distinguished Research Professor of Biomedicine, Medical University of South Carolina, 80 Barre Street, Charleston, South Carolina 29401, USA
- Dr I. Tagaya, Director, Department of Enteroviruses, National Institute of Health, Murayama Annex, Nakato, Musashimurayama, Tokyo 190-12, Japan

Secretary of Consultative Group

- Dr F.T. Perkins, Chief, Biological Standardization Unit, WHO, Geneva

Consultants

- Dr M.S. Balayan, Head, WHO Team for Special Studies in Virology, East African Virus Research Institute, P.O. Box 49, Entebbe, Uganda
- Dr S. Biberi-Moroeanu, State Institute for Sera and Vaccines, Splaiul Independentei 103, Bucharest 35, Romania
- Dr A.A. Combiescu, Head, Department of Enteroviruses, Institute of Microbiology, Parasitology and Epidemiology "Dr I. Cantacuzino", Splaiul Independentei 103, Bucharest 35, Romania
- Dr Bennett L. Elisberg, Director, Division of Pathology, Bureau of Biologics, Food and Drug Administration, Rockville, Maryland 20852, USA
- Dr F. Fornosi, Chief, Department of Virus Vaccine Control, National Institute of Hygiene, Gyali ut. 2-6, H-1966 Budapest IX, Hungary
- Dr C.L. Greening, Oficina Sanitaria Panamericana Zona II, Havre No. 30, 3^o y 4^o pisos, Colonia Juarez, Mexico 6, D.F., Mexico
- Dr D. Magrath, Division of Viral Products, National Institute for Biological Standards and Control, Holly Hill, Hampstead, London NW3 6RB, United Kingdom

* Dr H. Meyer, Bureau of Biologics, Food and Drug Administration, USA, was unable to attend and Dr L. Elisberg (see consultants) represented him.

Secretariat

Dr F.A. Assaad, Medical Officer, Virus Diseases Unit, WHO, Geneva

Dr P. Brès, Chief, Virus Diseases Unit, WHO, Geneva

Dr W.C. Cockburn, Director, Division of Communicable Diseases, WHO, Geneva

Observer

Dr de Mucha, Secretaria de Salubridad y Asistencia, Mexico, D.F., Mexico

AGENDA

Opening of the meeting by the Director-General

Election of a Chairman, Vice-Chairman and Rapporteur

1. WHO inquiry into acute persisting spinal paralysis
2. Report on collaborative studies on poliovirus strains isolated from cases temporally associated with the use of live poliovirus vaccine
3. The present situation in several countries in connection with production of vaccine
4. Distribution of type 2 and type 3 seed viruses
5. The laboratory tests of Pfizer and Russian type 3 vaccine
6. Report on a small-scale trial on Sabin type 3 vaccine prepared from second passage material on WI-38 cells of WHO seed lot (SO+1)
7. Report on the use of vaccine made from WHO type 3 seed (Glaxo seed pool 6) in Romania
8. Report on the status of collaborative studies of potency testing oral poliovirus vaccines
9. Report on the status of the original seed viruses of oral poliovirus vaccine
10. Report on the status of HEp-2 (Cincinatti) cells
11. Action to be taken to approve laboratories with a long record of production of vaccine
12. Approval of a new vaccine production technique in Mexico
13. Inhibitory substance in human saliva as a factor responsible for low efficacy of live poliovirus vaccine in tropics
14. The testing of neurovirulence of poliovirus vaccines in cynomolgus and rhesus monkeys
15. Stability of Sabin's type 2 vaccine strain

AN OUTLINE OF THE PUBLICATION ON THE
STUDY OF ACUTE PERSISTING SPINAL PARALYSIS

Introduction, including the

recommendation of the consultation on poliomyelitis in 1969

definitions

classifications

History of poliomyelitis in each country

History of vaccination in each country

Results

Classification of cases in each country and analysis by:

age and sex

seasonal distribution

interval between vaccination and onset of illness

laboratory findings

Vaccination history) mention briefly in narrative only

)

Marker tests) (refer to collaborative study)

Comparison between countries

Discussions

Conclusions

AN INTERNATIONAL COLLABORATIVE STUDY ON
TITRATION OF POLIOMYELITIS VIRUS

The lack of agreement on the virus content of poliomyelitis vaccines among manufacturers and control laboratories in several countries is still causing concern. It was agreed that an international collaborative assay would be helpful to resolve these difficulties.

The study would take place in two phases. The first phase would use monovalent vaccines of each of the virus types using a "study method" and a "local method" for titration. The steps to be taken would be the following:

1. The agreement on the details of the "study method".
2. The recording of the details of the "local method".
3. The procurement of monovalent virus suspensions (vaccines).
4. The agreement on the selection of reference preparations (monovalent).
5. The procurement of non cross-reactive monospecific antisera.
6. Agreement on the use of HEP-2 (Cincinnati) cells at an agreed passage level and monkey kidney cells.

Each monovalent virus and reference preparation would be titrated using both the "study method" and "local method", on each cell substrate and on each of three working days. The individual data should be reported to the WHO coordinator.

The monovalent vaccines and reference preparations would be titrated also in the presence of the heterotypic antisera at different dilutions on at least two working days using both titration methods and both cell substrates. The individual data should be reported to the WHO coordinator.

At this stage a report of the data and their analyses would be prepared and examined by the participants for:

- (a) agreement among laboratories on the most suitable method of titration, and
- (b) suitability of the monospecific sera for use in phase II of the study.

When agreement has been reached on (a) and (b), phase II of the study involving trivalent vaccine would start. The steps to be taken would be the following:

1. The agreement of the details of the titration method to be adopted.
2. The mixture of accurately measured quantities of the monovalent vaccines used in phase I of the study to make a trivalent vaccine.
3. The selection of several batches of trivalent vaccine.
4. Agreement on the use of HEP-2 (Cincinnati) cells and monkey kidney cells.
5. Agreement on the dilution of the selected monospecific sera to be used.

Each trivalent vaccine would be titrated on both cell substrates in the presence of the monospecific antibody on each of three working days. The individual data should be reported to the WHO coordinator.

A report of the data and their analyses would be prepared and examined by the participants. If agreement on a single titration method is reached it will be written into the Requirements for Poliomyelitis Vaccine (Oral) for the purposes of international agreement on the virus content of trivalent vaccine.