

## TRIAL OF A SUPPLEMENTAL DOSE OF FOUR POLIOVIRUS VACCINES

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**ABSTRACT**

**Background** The immunogenicity of oral poliovirus vaccine (OPV), particularly the type 3 component, is lower in infants in most developing countries than in infants in industrialized countries. We conducted a multicenter trial in Oman to evaluate the response to a supplemental dose of four poliovirus vaccine formulations.

**Methods** At nine months of age, infants were randomly assigned to receive inactivated-poliovirus vaccine (IPV), administered subcutaneously; trivalent OPV manufactured in the United States or in Europe; or monovalent type 3 OPV. Serum samples were collected at enrollment and 7 and 30 days later. All of the infants had previously received five doses of OPV.

**Results** We enrolled 1025 infants; 785 (76.6 percent) met all the study requirements. At enrollment, 96.8 percent of the infants were seropositive for poliovirus type 1, 98.0 percent for type 2, and 88.0 percent for type 3. At 30 days there were no significant increases in type 3 seroprevalence or in the median antibody titer in the groups of infants who received OPV. Among the recipients of IPV, type 3 seroprevalence increased from 87.8 percent at enrollment to 97.1 percent at 30 days ( $P < 0.001$ ), and the median antibody titer increased from 1:228 to 1:1448 or higher ( $P < 0.001$ ). The rapid initial increase in the antibody titer suggests a secondary immune response.

**Conclusions** A supplemental dose of IPV has excellent immunogenicity and leads to increases in the titer of antibodies against type 3 poliovirus, whereas supplemental doses of the oral vaccines do not have these effects. (N Engl J Med 2000;343:767-73.)

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**T**HE immunogenicity of oral poliovirus vaccine (OPV) is lower in infants in many developing countries than in infants in industrialized countries.<sup>1-5</sup> A review of studies in developing countries showed that the administration of three doses of OPV resulted in a median seroprevalence of 72 percent for poliovirus type 1, 95 percent for type 2, and 65 percent for type 3.<sup>6</sup> In contrast, three doses of OPV usually result in seroconversion in more than 95 percent of children in industrialized countries.<sup>7</sup> Several factors have been proposed to explain this difference.<sup>4,6,8-10</sup>

The type-specific antibody responses after vaccination with OPV are lower for poliovirus type 3 than for

types 1 and 2 in many developing countries,<sup>6</sup> including Oman, on the Arabian peninsula.<sup>11-13</sup> In 1990, a five-dose routine regimen of vaccination was introduced in Oman, with doses administered at birth, at 40 days, and at 3, 5, and 7 months.<sup>13-15</sup> In subsequent studies of seroconversion in Oman, the seroprevalence of poliovirus type 3 was 73 percent after four doses of OPV<sup>13</sup> and 80 percent after six doses.<sup>16</sup>

A number of approaches have been proposed to improve immunity against poliovirus type 3, including the administration of additional doses of OPV,<sup>17</sup> the administration of type-specific monovalent vaccines,<sup>18</sup> the use of trivalent OPV with an increased amount of type 3 virus,<sup>19,20</sup> the use of OPV manufactured in the United States,<sup>6</sup> and the substitution of all or some OPV doses with inactivated-poliovirus vaccine (IPV).<sup>21,22</sup>

At the nine-month visit for measles vaccination, we gave infants in Oman<sup>23</sup> a supplemental dose of one of four poliovirus vaccines in order to determine the most effective means of improving the immune responses to type 3 poliovirus and to determine the proportion of infants with secondary immune responses in the group of infants who were seronegative at nine months. The specific objectives of the study were, first, to compare humoral antibody responses (seroprevalence and antibody titer) after a single supplemental dose of IPV, trivalent OPV produced by a U.S. or European manufacturer, or monovalent type 3 OPV; and second, to characterize the serologic immune response as primary or secondary in previously seronegative infants. The study was conducted between August 1992 and April 1993 at four sites in Oman (Royal Hospital, Muscat; Seeb Health Center; and Ibra and Nizwa regional hospitals).

**METHODS****Study Design**

During the immunization visit at seven months, we informed parents or guardians about the study and asked them to participate if they lived within a one-hour drive of the study site. Healthy nine-month-old infants were eligible for enrollment if they had received five doses of OPV previously, with the last dose administered at

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least six weeks earlier. Infants were excluded if they had a medical condition requiring hospitalization, if they were known to be immunocompromised, or if they had a family history of an immunodeficiency disorder.

Voluntary, informed consent for participation in the study was obtained from the children's parents or guardians in accordance with the ethical principles enumerated in the Declaration of Helsinki and the additional requirements of local and national authorities. The study was approved by the Ministry of Health and the National Maternal–Child Health Committee, Muscat, Oman; the institutional review board of the Centers for Disease Control and Prevention, Atlanta; and the Secretariat Committee for Research Involving Human Subjects, World Health Organization (WHO), Geneva.

A standard questionnaire concerning demographic characteristics, medical and vaccination histories, family composition, and socioeconomic status was administered to each infant's parent or guardian. A short questionnaire about acute illnesses, including diarrhea, was administered during the follow-up visits at 7 and 30 days.

At each study site, the infants were randomly assigned to one of four vaccination groups with the use of blocks of 200 randomization numbers provided by the WHO. After a block of numbers had been used, another block of 200 numbers was provided. One group of infants received a subcutaneous dose of IPV (Imovax Polio, Pasteur Mérieux Serums et Vaccins, Lyons, France), formulated to contain 40 D antigen units of poliovirus type 1, 8 D antigen units of type 2, and 32 D antigen units of type 3. A second group received a dose of trivalent OPV (Orimune, Wyeth–Lederle Vaccines and Pediatrics, Pearl River, N.Y.) formulated to contain, on average,  $10^{6.5}$ ,  $10^{5.6}$ , and  $10^{6.3}$  median tissue-culture infective doses (TCID<sub>50</sub>) of Sabin poliovirus types 1, 2, and 3, respectively. The third group received a dose of trivalent OPV (SmithKline Beecham, Rixensart, Belgium) formulated to contain at least  $10^6$ ,  $10^5$ , and  $10^{5.8}$  TCID<sub>50</sub> of Sabin poliovirus types 1, 2, and 3, respectively. The fourth group received a dose of monovalent type 3 OPV (SmithKline Beecham) formulated to contain  $10^{5.8}$  TCID<sub>50</sub> of Sabin poliovirus type 3.

OPV was shipped on dry ice from the manufacturers to Oman. Samples from each lot of OPV, as well as randomly collected samples from the study sites, were sent to WHO collaborating centers for potency testing. Vaccine cold-chain–monitoring cards, which detect exposure to heat, were activated by the manufacturer before shipment and remained with the vaccine during shipment and storage.<sup>24</sup> All the lots of OPV were stored at  $-20^{\circ}\text{C}$ , with the temperature monitored twice daily; no breaks in the cold chain were recorded.

The potency of the OPV was estimated at two independent WHO collaborating centers according to standard WHO procedures.<sup>25</sup> The results are reported as the means of the values at the two laboratories. The laboratories determined the titer of each poliovirus type as well as the total viral titer; the stability of the total viral titer was also determined. Loss of stability (i.e., a decrease in the total titer per dose, expressed in TCID<sub>50</sub>) was assessed after 48 hours at  $37^{\circ}\text{C}$ . The antibody titers in the U.S. OPV were  $10^{6.15}$  for type 1,  $10^{5.31}$  for type 2,  $10^{5.77}$  for type 3, and  $10^{6.37}$  for total virus. The titers in the European OPV were  $10^{6.27}$  for type 1,  $10^{5.56}$  for type 2,  $10^{5.89}$  for type 3, and  $10^{6.36}$  for total virus. The antibody titer for type 3 in the monovalent vaccine was  $10^{6.12}$ . The mean loss of stability, expressed as a log<sub>10</sub> titer, was 0.71 for the U.S. OPV, 0.07 for the European OPV, and 0.16 for the monovalent type 3 OPV.

IPV was supplied in prefilled, single-dose syringes and shipped from the manufacturer to Oman at  $2^{\circ}$  to  $8^{\circ}\text{C}$  with cold-chain–monitoring cards and with monitors to detect freezing.<sup>24</sup> The vaccine was stored at  $2^{\circ}$  to  $8^{\circ}\text{C}$  with both types of temperature monitors; no cold-chain breaks were detected. Tests conducted by the manufacturer showed that the lot of vaccine contained 53 D antigen units of poliovirus type 1, 10 D antigen units of type 2, and 36 D antigen units of type 3 per dose. An independent WHO collaborating center confirmed the manufacturer's results.<sup>26</sup> Randomly selected field samples of IPV showed no decrease in potency.

Blood specimens were collected at enrollment and 7 and 30

days later. The serum was separated immediately, frozen, and stored at the study site at  $-20^{\circ}\text{C}$  pending shipment to the Centers for Disease Control and Prevention (CDC), in Atlanta. The specimens were tested by the CDC in triplicate with the use of modified neutralization assays for antibody to poliovirus types 1, 2, and 3. Serial dilutions of serum (from 1:8 to 1:1024) were incubated with 100 TCID<sub>50</sub> of poliovirus types 1, 2, and 3 at  $36^{\circ}\text{C}$  for three hours. HEp-2 (C) cells ( $1 \times 10^4$  to  $2 \times 10^4$ ) were then added to each well. The results of serologic tests are reported as titers of diluted serum that exhibited 50 percent neutralization.

Seropositivity was defined as a titer of 1:8 or higher.<sup>27</sup> A secondary immune response was defined as a titer of 1:8 or higher in serum obtained seven days after vaccination in infants with no detectable antibody ( $\leq 1:7$ ) at enrollment.

A follow-up study was performed when the infants were 15 months of age to assess the persistence of poliovirus neutralizing antibody and to measure indirectly the intestinal (mucosal) immunity to poliovirus type 3 induced by the three vaccines, with the use of a challenge dose of monovalent type 3 OPV (TCID<sub>50</sub>,  $10^{6.12}$ ). Blood specimens were collected at 15 months and 30 days later. Neutralizing antibody was measured by the CDC. Stool specimens were collected when the infants were 15 months of age (before the administration of the challenge dose of monovalent type 3 OPV) and 7 days later. Aliquots of stool were stored at  $-20^{\circ}\text{C}$  and shipped on dry ice to the Regional Virus Laboratory, Ruchill Hospital, Glasgow, United Kingdom. Stool specimens were examined for the presence of poliovirus according to standard procedures established by the WHO.<sup>28</sup>

### Statistical Analysis

After adjustment for the estimated proportion of seronegative infants (30 percent), we determined that a sample of at least 640 infants (160 in each of the four study groups) was needed to detect an increase in the seroprevalence of type 3 poliovirus that was at least 35 percent higher among the infants who received IPV or monovalent type 3 OPV than among those who received European OPV (with an alpha level of 0.05 and a beta level of 0.20 by a two-tailed test).

Statistical analyses were performed with the use of SAS software.<sup>29</sup> Comparisons of seroprevalence were performed with chi-square tests. Two-sided P values are reported. Because there were multiple comparisons among the four vaccination groups, a P value of 0.01 or less was considered to indicate statistical significance. Antibody titers were compared with the use of the Kruskal–Wallis nonparametric test.

## RESULTS

### Infants

A total of 1025 infants were enrolled in the study. Twenty-five infants (2.4 percent) were not seen at one or both visits during the 30-day period after vaccination. A total of 215 infants (21.5 percent) were excluded from the analysis because an insufficient quantity of serum was obtained at one or more visits. Complete data were thus available for 785 infants who participated in the initial study (76.6 percent) (Table 1). For the follow-up study, complete data were available for a total of 597 infants (i.e., data were available from all six visits, with a sufficient quantity of serum obtained at each visit).

### Immunogenicity at Nine Months

At enrollment, the seroprevalence of poliovirus types 1, 2, and 3 was 96.8, 98.0, and 88.0 percent, respectively; the corresponding median titers were 1:724, 1:1152, and 1:227 (Table 2). The seroprevalence did

**TABLE 1.** INFANTS PARTICIPATING IN THE INITIAL AND FOLLOW-UP STUDIES, ACCORDING TO THE STUDY SITE.

STUDY	ROYAL HOSPITAL	SEEB HEALTH CENTER	IBRA REGIONAL HOSPITAL	NIZWA REGIONAL HOSPITAL	TOTAL
	no. of infants				
Initial					
Visit 1 (at 9 mo)	275	200	275	275	1025
Visit 2 (7 days later)	267	199	271	271	1008
Visit 3 (30 days later)	262	198	271	269	1000
Complete data*	214	151	216	204	785
Follow-up					
Visit 4 (at 15 mo)	209	183	258	257	907
Visit 5 (7 days later)	208	183	257	256	904
Visit 6 (30 days later)	201	177	256	253	887
Complete data†	143	114	173	167	597

\*The numbers shown are the numbers of infants seen at all three visits, with sufficient serum obtained at each visit to test the sample in triplicate.

†The numbers shown are the numbers of infants seen at all six visits, with sufficient serum obtained at each visit to test the sample in triplicate.

not differ significantly according to the study site for type 1 (95.8 to 98.5 percent) or type 2 (97.1 to 98.6 percent). However, the seroprevalence of type 3 was lower in infants at the Ibra Regional Hospital (80.7 percent) than for those at the other three sites (89.4 to 92.1 percent,  $P=0.04$ ); the median antibody titer was also lower in Ibra (Table 2).

After randomization, the base-line seroprevalence ranged from 96.0 to 97.6 percent for type 1 poliovirus, from 96.6 to 99.4 percent for type 2, and from 83.8 to 93.2 percent for type 3 (Table 3). The base-line seroprevalence of type 3 in the group of infants who received U.S. OPV was significantly lower than that in the group who received monovalent type 3 OPV (83.8 percent vs. 93.2 percent,  $P=0.005$ ) but did not differ significantly from the seroprevalence among the infants who received IPV or European OPV.

At the 30-day visit, the seroprevalence did not differ significantly from the base-line values for poliovirus type 1 (an absolute increase of 1.5 to 2.4 percent) and type 2 (a change of -1.1 to 3.4 percent) in the three groups of infants who received vaccines containing all three viral serotypes. However, there were significant increases in the median antibody titer. The median titer of antibody against poliovirus type 1 increased from 1:576 at base line to 1:1448 or higher at 30 days in the group that received IPV, from 1:724 to 1:910 in the group that received U.S. OPV, and from 1:910 to 1:1152 in the group that received European OPV, with smaller increases in the titer of antibody against poliovirus type 2 in the IVP group and the U.S.-OPV group (from 1:1152 to  $\geq 1:1448$  and from 1:910 to 1:1152, respectively). Since serum was not tested at dilutions greater than 1:1028, the highest reportable titer was 1:1448 or higher.

**TABLE 2.** BASE-LINE SEROPREVALENCE OF AND MEDIAN SERUM ANTIBODY TITER FOR POLIOVIRUS TYPES 1, 2, AND 3, ACCORDING TO THE STUDY SITE.\*

POLIOVIRUS TYPE	ROYAL HOSPITAL (N=214)	SEEB HEALTH CENTER (N=151)	IBRA REGIONAL HOSPITAL (N=216)	NIZWA REGIONAL HOSPITAL (N=204)	TOTAL (N=785)
	no. of infants				
Type 1					
Seroprevalence (%)	96.7	96.0	95.8	98.5	96.8
Antibody titer	1:910	1:724	1:576	1:576	1:724
Type 2					
Seroprevalence (%)	98.6	98.0	97.1	98.0	98.0
Antibody titer	1:1152	1:1152	1:1152	1:1152	1:1152
Type 3					
Seroprevalence (%)	92.1	89.4	80.7†	90.2	88.0
Antibody titer	1:288	1:227	1:114	1:227	1:227

\*Seropositivity was defined as an antibody titer of 1:8 or higher.

†The seroprevalence of type 3 was significantly lower at the Ibra Regional Hospital than at the Royal Hospital ( $P=0.001$ ), Seeb Health Center ( $P=0.04$ ), or Nizwa Regional Hospital ( $P=0.01$ ).

The seroprevalence of poliovirus type 3 increased significantly in the group that received IVP (from 87.8 percent at base line to 97.1 percent at 30 days,  $P<0.001$ ); increased, but not significantly, in the group that received U.S. OPV (an absolute increase of 2.1 percent) and the group that received European OPV (0.6 percent); and decreased slightly in the group that received monovalent type 3 OPV (-1.0 percent). The median titer of antibody against poliovirus type 3 increased significantly, from 1:228 to 1:1448 or higher ( $P<0.001$ ), in the IPV group but did not change significantly in the other groups.

Among the infants who were seronegative at enrollment for type 1 (25 infants), type 2 (16), or type 3 (94), the rate of seroconversion at 30 days was 100 percent, 100 percent, and 76 percent, respectively, among those who received IPV and 37.5 percent, 33.3 percent, and 12.5 percent, respectively, among those who received U.S. OPV. In the group that received European OPV, the seroprevalence for types 1, 2, and 3 was 60 percent, less than 0 percent (99.4 percent at base line and 98.3 percent 30 days later), and 4.3 percent, respectively. In the group that received monovalent type 3 OPV, the seroprevalence was less than 0 percent (96.6 percent at base line and 96.1 percent 30 days later), 0 percent, and less than 0 percent (93.2 percent at base line and 92.2 percent 30 days later) for the three types, respectively.

#### Characterization of the Immune Response

In the group of infants who received IPV, the median titers of antibody against poliovirus types 1, 2, and 3 were already at the maximal level of detection at the seven-day visit (Table 3), suggesting that secondary immune responses had occurred. There were

**TABLE 3.** SEROPREVALENCE OF AND MEDIAN SERUM ANTIBODY TITER FOR POLIOVIRUS TYPES 1, 2, AND 3, ACCORDING TO THE STUDY GROUP.\*

POLIOVIRUS TYPE AND TIME OF TESTING	IPV (N=205)	U.S. OPV (N=198)	EUROPEAN OPV (N=177)	MONOVALENT TYPE 3 OPV (N=205)	TOTAL (N=785)
Type 1					
9-Mo visit					
Seroprevalence (%)	97.6	96.0	97.2	96.6	96.8
Antibody titer	1:576	1:724	1:910	1:910	1:724
7 Days later					
Seroprevalence (%)	99.5	97.0	97.7	95.6	97.5
Antibody titer	1:≥1448	1:724	1:1097	1:724	1:1152
30 Days later					
Seroprevalence (%)	100.0	97.5	98.9	96.1	98.1
Antibody titer	1:≥1448	1:910	1:1152	1:724	1:1152
Type 2					
9-Mo visit					
Seroprevalence (%)	96.6	97.0	99.4	99.0	98.0
Antibody titer	1:1152	1:910	1:≥1448	1:1152	1:1152
7 Days later					
Seroprevalence (%)	99.5	98.0	98.3	99.0	98.7
Antibody titer	1:≥1448	1:1152	1:≥1448	1:1152	1:≥1448
30 Days later					
Seroprevalence (%)	100.0	98.0	98.3	99.0	98.9
Antibody titer	1:≥1448	1:1152	1:≥1448	1:1152	1:≥1448
Type 3					
9-Mo visit					
Seroprevalence (%)	87.8	83.8†	87.0	93.2	88.0
Antibody titer	1:228	1:181	1:228	1:228	1:228
7 Days later					
Seroprevalence (%)	97.6	83.8	86.4	91.7	90.1
Antibody titer	1:≥1448‡	1:181	1:288	1:228	1:455
30 Days later					
Seroprevalence (%)	97.1‡	85.9	87.6	92.2	90.8
Antibody titer	1:≥1448	1:181	1:288	1:288	1:576

\*Seropositivity was defined as an antibody titer of 1:8 or higher. IPV denotes inactivated-poliovirus vaccine, and OPV oral poliovirus vaccine.

†P=0.005 for the comparison with the base-line value in the monovalent type 3 OPV group.

‡P<0.001 for the comparison with the base-line value in the IPV group.

less dramatic increases in the titers of antibody against types 1 and 2 in the group that received U.S. OPV and in the titer of antibody against type 1 in the group that received European OPV.

#### Persistence of Neutralizing Antibody

The follow-up study included 597 infants — those for whom complete data were available with valid serologic results (Table 1). In the group of infants who received monovalent type 3 OPV, the median antibody titer for each poliovirus type declined until the visit at 15 months (when a challenge dose was administered). Among the infants who received U.S. or European OPV, the titers of antibody against poliovirus types 1 and 2 increased, whereas the titer of antibody against type 3 gradually decreased. In the group of infants who received IPV, antibody titers for all three poliovirus types were at the highest detectable level (≥1:1448) during the first 30 days. The titer of antibody against poliovirus type 3 remained significantly higher in the group of infants who received

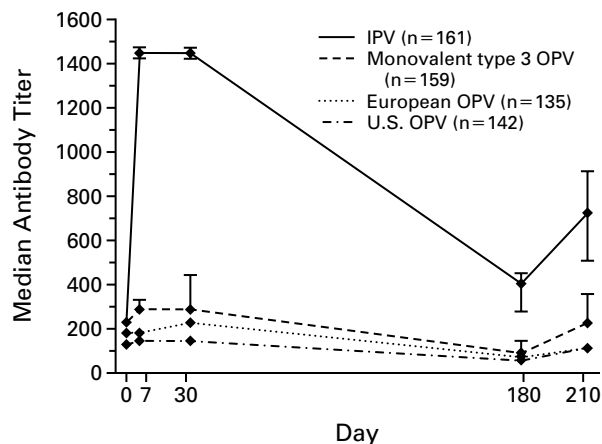
IPV than in the other three groups throughout the study period (Fig. 1).

#### Excretion of Challenge Virus

Mucosal immunity was assessed by administering a challenge dose of poliovirus type 3 (monovalent type 3 OPV) at the 15-month visit. Stool specimens were collected immediately before the challenge and seven days afterward. Children who excreted poliovirus just before the challenge were excluded from the post-challenge analysis. Overall, 33.6 percent of the infants excreted virus, including poliovirus type 3 (13.2 percent), nonpoliovirus enterovirus (14.5 percent), and adenovirus (4.4 percent) (Table 4). There were no significant differences in the rate of excretion of poliovirus type 3 among the study groups.

#### DISCUSSION

Immunity to poliovirus type 3 is suboptimal in infants in many developing countries. Our multicenter study in Oman was designed to determine whether



**Figure 1.** Median Serum Titers of Poliovirus Type 3 Antibody over Time in Infants in Oman Who Were Given a Supplemental Dose of Poliovirus Vaccine at Nine Months of Age, According to the Type of Vaccine.

Only the 597 infants for whom valid serologic results were available for all six visits were included in the analysis. The I bars indicate 95 percent confidence intervals for median serum antibody titers in the group of infants who received inactivated-poliovirus vaccine (IPV) and upper 95 percent confidence limits in the group that received monovalent type 3 oral poliovirus vaccine (OPV). The antibody titer was 1:1448 or higher in 116 of the infants in the IPV group at 7 days and in 117 at 30 days. The 95 percent confidence intervals for the median titers in the IPV group at 7 and 30 days were therefore equal to the point estimates (i.e.,  $\geq 1:1448$ ).

a supplemental dose of one of four poliovirus vaccines would improve immunity. Several of our findings have implications for the global initiative to eradicate polio. We found that monovalent type 3 OPV was no more effective than trivalent OPV in inducing a type-specific antibody response. The immunogenicity of U.S. OPV appeared to be similar to that of European OPV; however, IPV was significantly more immunogenic than either OPV or monovalent type 3 OPV and was associated with a particularly high seroprevalence and high median titer of antibody against poliovirus type 3. The majority of infants who were initially seronegative for poliovirus type 3 had secondary immune responses to the supplemental dose of IPV.

Monovalent vaccines are more effective in inducing a primary immune response than trivalent OPV, which requires a balanced formulation with a high level of potency for each poliovirus type and additional doses to match the performance of monovalent OPV.<sup>30</sup> Many reports from the late 1950s and early 1960s suggest that one dose of monovalent OPV induces a type-specific seroconversion level higher than 90 percent.<sup>31-35</sup> We gave monovalent type 3 OPV to infants who had received five previous doses of OPV. Our findings suggest that in OPV-vaccinated infants, most of whom have received multiple doses of OPV,

**TABLE 4.** EXCRETION OF VIRUS IN STOOL SPECIMENS OBTAINED AFTER A CHALLENGE DOSE OF POLIOVIRUS TYPE 3.

VIRUS	ROYAL HOSPITAL (N=203)	SEEB HEALTH CENTER (N=172)	IBRA REGIONAL HOSPITAL (N=266)	NIZWA REGIONAL HOSPITAL (N=254)	TOTAL (N=895)
Poliovirus type 1	0	1 (<1)	0	1 (<1)	2 (<1)
Poliovirus type 2	0	0	2 (<1)	10 (4)	12 (1)
Poliovirus type 3	30 (15)	33 (19)	32 (12)	23 (9)	118 (13)
Nonpoliovirus enterovirus	16 (8)	19 (11)	47 (18)	48 (19)	130 (15)
Adenovirus	15 (7)	8 (5)	7 (3)	9 (4)	39 (4)
None	142 (70)	111 (65)	178 (67)	163 (64)	594 (66)

monovalent vaccines offer no advantage over OPV in raising the level of immunity.

Some investigators have suggested that the OPV produced in the United States is more immunogenic than the OPV produced elsewhere, because it is formulated to contain more virus.<sup>6</sup> U.S. OPV is administered in single-dose dispensers with a larger volume (0.5 ml) than the standard dose of OPV used in other countries (0.1 ml). Furthermore, U.S. OPV is stabilized with sorbitol, whereas European OPV is stabilized with magnesium chloride. In our study, however, independent testing showed no differences in the potency of the two OPV formulations. In addition, there were no differences in their efficacy either in inducing a type-specific antibody response or in raising antibody titers in infants with detectable antibody levels.

The efficacy of IPV, a parenteral vaccine, was not affected by the mucosal immunity induced by previous doses of OPV. We found that a booster dose of IPV provided excellent immunogenicity in infants in a developing country, confirming the results of other studies.<sup>22,36</sup> IPV may be less immunogenic in the first several months of life, when the infant still has maternal antibody, than later in infancy.<sup>21</sup> Thus, in developing countries, routine use of IPV in early infancy may not be warranted, but a single dose of IPV given later in the first year of life (when maternal antibody has disappeared) could substantially increase the level of immunity to all three types of poliovirus, and especially type 3.<sup>21</sup>

Among the infants in our study who had no detectable antibody at base line, all those who received IPV had secondary responses (i.e., detectable antibody seven days after vaccination). This is an unexpected finding, and it suggests that priming had already been

induced by the initial series of vaccinations in these infants but that they either had no antibody or had titers below the cutoff level of 1:8. The secondary responses are consistent with the results in the entire group of infants who received OPV, in whom a supplemental dose of OPV did not result in significant increases in seroprevalence and resulted in only moderately increased antibody titers.

We assessed mucosal immunity by administering a challenge dose of monovalent type 3 OPV six months after the supplemental dose of vaccine had been administered. In previous studies, the challenge virus was poliovirus type 1<sup>21,37-40</sup> or trivalent OPV,<sup>41</sup> and the challenge dose was administered within one to three months after the last dose of vaccine. After the administration of a challenge dose of 10<sup>6.12</sup> TCID<sub>50</sub> of monovalent type 3 OPV, there were no significant differences in the excretion of poliovirus type 3 among our four study groups.

Our study had limitations. Because we did not assess mucosal immunity directly, we can only make inferences about mucosal immunity from serologic and excretion data, and the results should therefore be interpreted with caution. Previous reports suggest that priming occurs after vaccination, even in the absence of circulating antibody.<sup>42</sup> We do not know the sensitivity or specificity of our definition of a secondary immune response.

The results of our study have implications for the initiative to eradicate poliomyelitis throughout the world by the end of this year or as soon as possible thereafter. The eradication strategies, including the achievement of a high level of routine immunization coverage, the administration of supplemental doses of vaccine during national immunization days to reduce widespread circulation of poliovirus, house-to-house vaccination campaigns when the circulation of virus has become focal, and the establishment of a sensitive system of surveillance for poliovirus, appear to be successful.<sup>43,44</sup> The results of our study support the use of a supplemental dose of IPV at nine months of age after a routine primary series of vaccinations with OPV in infants in developing countries, particularly in areas where vaccination coverage is high (>90 percent of infants). However, the cost effectiveness of this strategy must be determined before it can be adopted as a policy. Once poliovirus has been eliminated in a country or region, the use of IPV may provide a benefit similar to that of national immunization days, in terms of individual protection, and thus may help maintain a high level of immunity in the population until programs of poliovirus vaccination can be terminated throughout the world.

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## REFERENCES

- Swartz TA, Skalska P, Gerichter CG, Cockburn WC. Routine administration of oral polio vaccine in a subtropical area: factors possibly affecting sero-conversion rates. *J Hyg (Lond)* 1972;70:719-26.
- John TJ. Problems with oral poliovaccine in India. *Indian Pediatr* 1972; 9:252-6.
- John TJ, Jayabal P. Oral polio vaccination of children in the tropics. I. The poor sero-conversion rates and the absence of viral interference. *Am J Epidemiol* 1972;96:263-9.
- Domök I, Balayan MS, Fayinka OA, Skrtic N, Soneji AD, Harland PS. Factors affecting the efficacy of live poliovirus vaccine in warm climates: efficacy of type 1 Sabin vaccine administered together with anti-human gamma-globulin horse serum to breast-fed and artificially fed infants in Uganda. *Bull World Health Organ* 1973;51:33-47.
- Chopra K, Kundu S, Chowdury DS. Antibody response of infants in tropics to five doses of oral polio vaccine. *J Trop Pediatr* 1989;35:19-23.
- Patriarca PA, Wright PF, John TJ. Factors affecting the immunogenicity of oral poliovirus vaccine in developing countries: review. *Rev Infect Dis* 1991;13:926-39.
- McBean AM, Thoms ML, Albrecht P, Cuthie JC, Bernier R. Serologic response to oral polio vaccine and enhanced-potency inactivated polio vaccines. *Am J Epidemiol* 1988;128:615-28.
- Sabin AB, Ramos-Alvarez M, Alvarez-Amezquita J, et al. Live, orally given poliovirus vaccine: effects of rapid mass immunization on population under conditions of massive enteric infection with other viruses. *Bull World Health Organ* 1999;77:196-201.
- Posey DL, Linkins RW, Oliveria MJ, Monteiro D, Patriarca PA. The effect of diarrhea on oral poliovirus vaccine failure in Brazil. *J Infect Dis* 1997;175:Suppl 1:S258-S263.
- Myaux JA, Unicomb L, Besser RE, et al. Effect of diarrhea on the humoral response to oral polio vaccination. *Pediatr Infect Dis J* 1996;15:204-9.
- Bernkopf H, Medalie J, Yetkutiel M. Antibodies to poliomyelitis virus and socioeconomic factors influencing their frequency in children in Israel. *Am J Trop Med Hyg* 1957;6:697-703.
- Pal SR, Banerjee G, Aikat BK. Serological investigation on endemicity of poliomyelitis in Calcutta and neighboring rural areas. *Indian J Med Res* 1966;54:507-11.
- Sutter RW, Patriarca PA, Suleiman AJM, et al. Paralytic poliomyelitis in Oman: association between regional differences in attack rate and variations in antibody responses to oral poliovirus vaccine. *Int J Epidemiol* 1993;22:936-44.
- Robertson SE, Suleiman AJM, Mehta FR, al-Dhahry SHS, el-Bualy MS. Poliomyelitis in Oman: acute flaccid paralysis surveillance leading to early detection and rapid response to a type 3 outbreak. *Bull World Health Organ* 1994;72:907-14.
- Ministry of Health. Progress towards poliomyelitis eradication — Oman acute flaccid paralysis surveillance (AFP) 1990-1995. Community Health & Disease Surveillance Newsletter. Vol. 4. No. 4. 1995:1-5. (Muscat, Oman: Sultanate of Oman.)
- Sutter RW, Suleiman AJM, Malankar PG, et al. Sequential use of inactivated poliovirus vaccine followed by oral poliovirus vaccine in Oman. *J Infect Dis* 1997;175:Suppl 1:S235-S240.
- Patriarca PA, Linkins RW, Sutter RW, Orenstein WA. Optimal schedule for the administration of oral poliovirus vaccine. In: Kurstak E, ed. Measles and poliomyelitis: vaccines, immunization, and control. New York, Springer-Verlag, 1993:303-13.
- Schoub BD, Johnson S, McAnerney J, Gilbertson L, Klaasen KIM, Reinach SG. Monovalent neonatal polio immunization — a strategy for the developing world. *J Infect Dis* 1988;157:836-9.
- Patriarca PA, Laender F, Palmeira G, et al. Randomised trial of alternative formulations of oral poliovaccine in Brazil. *Lancet* 1988;1:429-33.

20. Factors affecting the immunogenicity of oral poliovirus vaccine: a prospective evaluation in Brazil and The Gambia. *J Infect Dis* 1995;171:1097-106.
21. Combined immunization of infants with oral and inactivated poliovirus vaccines: results of a randomized trial in The Gambia, Oman, and Thailand. *J Infect Dis* 1997;175:Suppl 1:S215-S227.
22. Moriniere BJ, van Loon FPL, Rhodes PH, et al. Immunogenicity of a supplemental dose of oral versus inactivated poliovirus vaccine. *Lancet* 1993;341:1545-50.
23. Expanded Programme on Immunization. Immunization policy. Geneva: World Health Organization, 1996. (WHO/EPI/GEN/95.03 REV 1.)
24. *Idem*. Product information sheets. Geneva: World Health Organization, 1997. (WHO/EPI/LHIS/97.01.)
25. Vaccine supply and quality: manual of laboratory methods. Geneva: World Health Organization, 1997. (WHO/VSQ/97.04.)
26. Minor P. Summary report of a meeting on the estimation of the potency of inactivated poliovaccine. *Biologicals* 1990;18:243-4.
27. Albrecht P, Enterline JC, Boone EJ, Klutch MJ. Poliovirus and polio antibody assay in HEp-2 and Vero cell cultures. *J Biol Stand* 1983;11:91-7.
28. Expanded Programme on Immunization, Division of Communicable Diseases. Manual for the virological investigation of poliomyelitis. Geneva: World Health Organization, 1990. (WHO/EPI/CDS/POLIO 90.1.)
29. SAS/STAT user's guide, version 6.4. 4th ed. Vol. 1. Cary, N.C.: SAS Institute, 1989.
30. Robertson HE, Acker MS, Dillenberg HO, et al. Community-wide use of a "balanced" trivalent oral poliovirus vaccine (Sabin): a report of the 1961 trial at Prince Albert, Saskatchewan. *Can J Public Health* 1962;53:179-91.
31. Horwitz A, Martins da Silva M, Bica AN. Large-scale field studies with live attenuated poliovirus vaccines in the Americas. In: Poliomyelitis: papers and discussions presented at the 5th International Poliomyelitis Conference, Copenhagen, Denmark, July 26-28, 1960. Philadelphia: J.B. Lippincott, 1961:221-7.
32. Kurnosova LM, Zhilova GP. Immunologic changes in the blood of children inoculated with live poliomyelitis vaccine. In: Live vaccine against poliomyelitis. Leningrad, Russia: Institute of Experimental Medicine of the USSR Academy of Medical Sciences, 1960.
33. Voroshilova MK. Influence of dose and schedule of oral immunization of people with live poliovirus vaccine on antibody response. In: Poliomyelitis: papers and discussions presented at the 5th International Poliomyelitis Conference, Copenhagen, Denmark, July 26-28, 1960. Philadelphia: J.B. Lippincott, 1961:296-303.
34. Verlinde JD, Wilterdink JB. A small-scale trial on vaccination and revaccination with live attenuated polioviruses in the Netherlands. In: First International Conference on Live Poliovirus Vaccines. Washington, D.C.: Pan American Health Organization, 1959:355-66. (Scientific publication no. 44.)
35. Cox HR, Cabasso VJ, Markham PS, et al. Immunologic response to trivalent oral poliomyelitis vaccine. In: First International Conference on Live Poliovirus Vaccines. Washington, D.C.: Pan American Health Organization, 1959:229-48. (Scientific publication no. 44.)
36. Plotkin SA, Murdin A, Vidor E. Inactivated polio vaccine. In: Plotkin SA, Orenstein WA, eds. Vaccines. 3rd ed. Philadelphia: W.B. Saunders, 1999:345-63.
37. Onorato IM, Modlin JF, McBean AM, et al. Mucosal immunity induced by enhanced-potency inactivated and oral polio vaccines. *J Infect Dis* 1991;163:1-6.
38. Glezen WP, Lamb GA, Belden EA, Chin TDY. Quantitative relationship of preexisting homotypic antibodies to the excretion of attenuated poliovirus type 1. *Am J Epidemiol* 1966;53:224-37.
39. Ghendon YZ, Sanakoyeva II. Comparison of the resistance of the intestinal tract to poliomyelitis (Sabin's strains) in persons after naturally and experimentally acquired immunity. *Acta Virol* 1961;5:265-73.
40. Kok PW, Leeuwenberg J, Tukei P, et al. Serological and virological assessment of oral and inactivated poliovirus vaccines in a rural population in Kenya. *Bull World Health Organ* 1992;70:93-103.
41. Modlin JF, Halsey NA, Thoms ML, Meschievitz CK, Patriarca PA. Humoral and mucosal immunity in infants induced by three sequential inactivated poliovirus vaccine-live attenuated oral poliovirus vaccine immunization schedules. *J Infect Dis* 1997;175:Suppl 1:S228-S234.
42. Salk J, Stoeckel P, van Wezel AL, Lapinleimu K, van Steenis G. Antigen content of inactivated poliovirus vaccine for use in a one- or two-dose regimen. *Ann Clin Res* 1982;14:204-12.
43. Hull HF, Ward NA, Hull BP, Milstien JB, de Quadros C. Paralytic poliomyelitis: seasoned strategies, disappearing disease. *Lancet* 1994;343:1331-7.
44. Progress toward global poliomyelitis eradication — 1997-1998. *MMWR Morb Mortal Wkly Rep* 1999;48:416-21.