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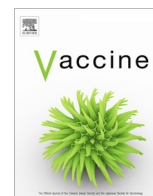
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Evaluation of vaccine derived poliovirus type 2 outbreak response options: A randomized controlled trial, Karachi, Pakistan



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ABSTRACT

Background: Outbreaks of circulating vaccine derived polioviruses type 2 (cVDPV2) remain a risk to poliovirus eradication in an era without live poliovirus vaccine containing type 2 in routine immunization. We evaluated existing outbreak response strategies recommended by the World Health Organization (WHO) for control of cVDPV2 outbreaks.

Methods: Seronegative children for poliovirus type 2 (PV2) at 22 weeks of life were assigned to one of four study groups and received respectively (1) one dose of trivalent oral poliovirus vaccine (tOPV); (2) monovalent OPV 2 (mOPV2); (3) tOPV together with a dose of inactivated poliovirus vaccine (IPV); or (4) mOPV2 with monovalent high-potency IPV type 2. Stool and blood samples were collected and assessed for presence of PV2 (stool) and anti-polio antibodies (sera).

Results: We analyzed data from 265 children seronegative for PV2. Seroconversion to PV2 was achieved in 48, 76, 98 and 100% in Groups 1–4 respectively. mOPV2 was more immunogenic than tOPV alone ($p < 0.001$); and OPV in combination with IPV was more immunogenic than OPV alone ($p < 0.001$). There were 33%, 67%, 20% and 43% PV2 excretors in Groups 1–4 respectively. mOPV2 resulted in more prevalent shedding of PV2 than when tOPV was used ($p < 0.001$); and tOPV together with IPV resulted in lower excretion of PV2 than tOPV alone ($p = 0.046$).

Conclusion: mOPV2 was a more potent vaccine than tOPV. Adding IPV to OPV improved immunological response; adding IPV also seemed to have shortened the duration of PV2 shedding. mIPV2 did not provide measurable improvement of immune response when compared to conventional IPV. WHO recommendation to use mOPV2 as a vaccine of first choice in cVDPV2 outbreak response was supported by our findings. Clinical Trial registry number: NCT02189811.

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1. Background

In 2017, there were 22 reported cases of poliomyelitis caused by wild poliovirus type 1 (WPV1) detected in two remaining endemic countries (Afghanistan and Pakistan) [1]. WPV2 is considered eradicated and WPV3 was last detected in 2012, in Nigeria [2,3].

Complete poliovirus eradication, however, requires the disappearance of not only WPVs but of all polioviruses from human

populations, including those resulting from use of oral poliovirus vaccine (OPV). The Polio Eradication & Endgame Strategic Plan 2013–2018 provides a framework for interruption of WPV transmission in remaining endemic foci and lays out plan for the new polio endgame, which includes the sequential withdrawal of Sabin virus strains contained in OPV vaccine, starting with type 2, and the introduction of inactivated poliovirus vaccine (IPV), for risk mitigation purposes [4].

The switch from trivalent OPV (tOPV) to bivalent OPV (bOPV) without type 2 poliovirus (PV2) was conducted in a globally synchronized manner in April 2016. As of May 2016, there were no countries still using type 2-containing OPV in routine immunization; however, the World Health Organization (WHO) maintains

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a stock of monovalent type 2 OPV (mOPV2) reserved for outbreak response in case of outbreaks of type 2 circulating vaccine derived poliovirus (cVDPV2) or accidental release of WPV2 in the post-switch era [5]. The use of mOPV2 must be authorized by the Director General of WHO. In addition to mOPV2, monovalent inactivated poliovirus vaccine type 2 (mIPV2) with 4-times higher PV2 antigenic potency than standard IPV has been evaluated in clinical trials and found to be safe and immunogenic; however, mIPV2 has not been licensed, is considered experimental, and is not part of the outbreak response toolkit recommended by WHO [6,7].

As per WHO's Poliovirus Outbreak Response Guidelines, bOPV and IPV or mOPV2 and IPV are tools for control of WPV1 or cVDPV2 outbreaks respectively [8]. Since the switch from tOPV to bOPV in April 2016 until December 2017, there were 44 separate incidents when VDPV2 was detected either in children or from environmental samples [1]. In 12/44 incidents, mOPV2 was authorized to be used to control the outbreak and in 7/44 incidents, IPV was used [1,9]. The decision regarding what outbreak response tools should be deployed is based on the assessment of risk of poliovirus circulation, and risk of exportation to other countries.

In the post-switch era, the risk of emergence and spread of cVDPV2 is two-fold, from unrecognized foci seeded before the tOPV to bOPV switch such as long-term excretors among immunodeficient individuals, or from post-switch use of PV2-containing live vaccine such as mOPV2 or left-over tOPV.

To assess the available tools for cVDPV2 outbreak response, we analyzed a subset of data obtained from a larger study that was conducted by the same investigators as this analysis in Pakistan and was entitled "Immunogenicity of Different Routine Poliovirus Vaccination Schedules: a Randomized Controlled Trial". That study had two objectives: to assess the immunogenicity of different routine immunization schedules; and to assess different poliovirus outbreak response strategies. Here, we present the data obtained to assess the polio outbreak response strategies.

2. Methods

The study was conducted in four low-income areas in and around Karachi (4 peri-urban, contiguous coastal villages: Rehri Goth, Bhains Colony, Ali Akber Shah and Ibrahim Hydri) where the Aga Khan University's Department of Paediatrics and Child Health has a well-established demographic surveillance system (DSS) which captures population size, pregnancies and births. The population of the study area according to the last census from 2015 was 294,171. Each area has a Primary Health Center (PHC) operated by the Department of Paediatrics and Child Health Research Program of the Aga Khan University, which also provides Expanded Programme on Immunizations (EPI) services.

2.1. Selection of study participants

Expectant women were approached at home during pregnancy or immediately after delivery by health center staff and information about the trial and an invitation to participate in the study were given. Inclusion criteria included healthy newborns with birth weight ≥ 2.0 kg, informed consent from parent or guardian, and residence in the service area of the study clinic, with no plans to move during the study period. Newborns who had any clinical sign of illness using the WHO Integrated Management of Neonatal and Childhood Illness assessment tool, required hospitalization, or were at risk of immunodeficiency (through a family history screen) were excluded from the trial.

After enrollment into "Immunogenicity of Different Routine Poliovirus Vaccination Schedules: a Randomized Controlled Trial", children were randomized to receive one of four different polio pri-

mary immunization schedules, with one dose of poliovirus vaccine administered at birth, 6, 10, and 14 weeks of age. The four schedules were 1: IPV, IPV, IPV, IPV; 2: bOPV, bOPV, bOPV, bOPV; 3: bOPV, bOPV, bOPV, bOPV+IPV; 4: tOPV, tOPV, tOPV, tOPV [10].

Some of the enrolled children had remained seronegative for poliovirus type 2 (PV2) at 22 weeks of age; and these children formed the study set for this analysis. We divided the seronegative children into four different groups and administered poliovirus vaccines to them at 22 weeks of age: in group 1 they received one dose of tOPV; group 2 received mOPV2; group 3 received tOPV together with IPV; and group 4 received mOPV2 together with mIPV2 (Table 1). The allocation to OPV alone or OPV+IPV groups was random; however, the allocation to tOPV or mOPV2 groups was not: the trial started while permission to use mOPV2 and mIPV2 was being processed and therefore those children enrolled before the permission was granted (in November 2015) had been randomized into tOPV or tOPV+IPV groups (Group 1 or 3); and those children enrolled after November 2015 had been randomized into mOPV2 or mOPV2+mIPV2 groups (Group 2 or 4).

2.2. Study Procedures, and definitions

Peripheral blood (2 mL) was collected at 22, 23 and 26 weeks of age. Blood specimens collected at the sites were allowed to clot, centrifuged to separate serum, and transported to the Infectious Disease Research Laboratory (IDRL) at the Aga Khan University where they were stored at -20°C until shipment to the Centers for Disease Control and Prevention (CDC), Atlanta, Georgia, USA, where the sera were tested for presence of poliovirus neutralizing antibodies using standard neutralization assays [11].

Seropositivity was defined as reciprocal titers of poliovirus neutralizing antibodies ≥ 8 ; seroconversion was defined as a change from seronegative to seropositive (from reciprocal titer of <8 to ≥ 8).

PV2 priming in children who had previously received IPV but remained seronegative at 22 weeks of age was inferred for children who had an anamnestic immune response (seroconversion) within 7 days of vaccination.

Stool specimens were collected at the primary care clinic or at children's homes on week 22 (before study vaccine administration) and on week 23 and were stored at IDRL at $+4^{\circ}\text{C}$ for a maximum of one week until shipment to WHO Regional Reference Laboratory for polio at the National Institute of Health in Islamabad, Pakistan, where they were tested for the presence of poliovirus using standard poliovirus detection methodology [12]. Presence or absence of PV2 in stool samples was reported.

Adverse events following vaccination were identified by site investigators and reviewed by the principal investigator. Children were observed for 30 min following the administration of the vaccine for immediate adverse events; parents were instructed to immediately report back to the health centers if adverse events occurred after the initial observation period. Serious adverse events were reported for review to the Data and Safety Monitoring Board and the Ethical Review Committee.

IPV and mIPV2 were produced by Bilthoven Biologicals B.V., the Netherlands, and presented in 1-dose vials (0.5 mL), tOPV(20-dose vials) and mOPV2(20-dose vials) were produced by Sanofi Pasteur in vials for oral administration.

2.3. Sample size and analysis

Target sample size for each arm in the "Immunogenicity of Different Routine Poliovirus Vaccination Schedules: a Randomized Controlled Trial" study was calculated to be 190 newborns with a minimum analyzable sample size of 110 per arm, accepting $\alpha = 0.05$ and power = 80%, and assuming at least 20% difference

Table 1
Description of study groups and biological samples obtained.

Study group	Birth	0–14 wks.	22 wks.	23 wks.	26 wks.
1	Enrolment	Different primary polio immunization schedules	tOPV		
2			mOPV2		
3			tOPV+IPV		
4			mOPV2+mIPV2		
Samples collected			Blood Stool	Blood Stool	Blood

in seroconversion between arms. The study population for the evaluation of polio outbreak tools consisted of children who had been enrolled in this study and who remained seronegative for PV2 at 22 weeks of age; therefore, specific sample size calculation was impossible to perform.

Data was analyzed using STATA version 11. The proportion of seroconversion in different study arms was compared by χ^2 test for quantitative variables. P-value was calculated to assess difference between study arms. K-sample equality of median test was performed to compare the median titers across the study arms and 95% confidence intervals for median titers were calculated using bootstrap methods.

The study was approved by the Ethical Review Committee of the Aga Khan University, the National Bioethics Committee of Pakistan, and the Ethical Review Committee of the World Health Organization, Geneva. All activities followed the guidelines of Good Clinical Practice; the trial protocol was registered at ClinicalTrials.gov with identifier NCT02189811. The World Health Organization assisted in study design, trial implementation and monitoring, and contributed to writing of the report. The Aga Khan University conducted the trial. The National Institute of Health, Pakistan, tested stool samples and CDC tested the sera.

3. Results

A total of 1481 newborns were assessed for eligibility for the larger study and 900 newborns were enrolled in the “Immuno-genicity of Different Routine Poliovirus Vaccination Schedules: a Randomized Controlled Trial”. Of these, there were 265/900 (29.4%) who remained seronegative at 22 weeks of age for PV2 [10]; and these 265 children formed our study population.

There were 145, 53, 52, and 15 children assigned to groups 1–4 respectively (Fig. 1). Baseline demographic indicators are shown in Table 2; there were no significant differences between groups in any of the indicators. There was higher number of children assigned to Group 1 than to the other groups because these were the children that had received bOPV vaccine in the larger study and therefore remained mostly seronegative for PV2.

We assessed seroconversion for PV2 one and four weeks after administration of the study vaccines (Table 3). mOPV2 alone induced seroconversion significantly more often than tOPV alone (76 vs 48%, $p < 0.001$); mOPV2+mIPV2, however, did not provide significantly higher seroconversion than tOPV+IPV (100 vs 98%, $p = 0.6$). IPV in combination with OPV induced significantly higher seroconversion to type 2 than OPV alone ($p < 0.001$), regardless of whether mIPV2 or IPV was used. Antibody titers were higher when mOPV2 was used compared with tOPV ($p < 0.001$) and when mOPV2+mIPV2 was used compared with tOPV+IPV ($p < 0.001$).

IPV priming was assessed in children who had received prior IPV and compared with those who had not. Among children who seroconverted for PV2 between 22 and 26 weeks of age there were 101/164 (62%) who had received at least one IPV dose before 22 weeks of age but had not seroconverted by 22 weeks of age. There was no statistical difference in the proportion of subjects who seroconverted one week after OPV administration between prior-IPV

recipients and non-recipients (44 vs 56% for tOPV; and 87 vs 71% for mOPV2, $p > 0.05$). There were no children who had not received prior IPV in groups 3 and 4; all children in these two groups seroconverted within 1 week of vaccines administration (Fig. 2).

Excretion of PV2 in stool was assessed one week after vaccine administration in those who had no PV2 isolated in their stools on the day of vaccine administration (2/265 [1%] were excluded: one child in group 1 and one child in group 2). There were 33%, 67%, 20% and 43% PV2 excretors in Groups 1–4 respectively (Fig. 3). mOPV2 resulted in more prevalent shedding of PV2 than when tOPV was used ($p < 0.001$) and tOPV together with IPV resulted in lower excretion of PV2 than tOPV alone ($p = 0.046$). We stratified the analysis of PV2 excretion by primary immunization schedule the children had previously received, however, no significant differences in PV2 excretion were found between the strata.

No severe adverse events linked to administration of the study vaccines were reported.

4. Discussion

We demonstrated that mOPV2 is more immunogenic than tOPV and that IPV in combination with OPV is more immunogenic than OPV alone. In our study, one dose of mOPV2 administered at 22 weeks of age resulted in lower seroconversion than previously reported (communication with CDC). We were unable to assess whether mIPV2 provided better immunological response than conventional IPV because the combination of tOPV+IPV was already immunogenic in 98% of children; the increase from 98% to 100% (provided by the combination of mOPV2+mIPV2) did not reach statistical significance.

Previous data showed that priming after one IPV dose was common and assumed to provide protection against paralytic poliomyelitis in children who remain seronegative after past IPV administration. In one study in Cuba, priming after one IPV dose was estimated at 98% [13]. In our study we could not measure priming in the same way: there were no children in the IPV groups who had not received at least one prior IPV dose. However, seronegative children who had received at least one IPV dose and were subsequently vaccinated with OPV (tOPV or mOPV2) did not exhibit more frequent anamnestic immune response than children who had not received any prior IPV. It is therefore possible that the priming observed in Cuba occurs only when the subsequent poliovirus vaccine is IPV [13]. The role of priming in protection against poliomyelitis needs to be further evaluated. mOPV2 induced more frequent shedding of PV2 than did tOPV; IPV in combination with OPV resulted in lower proportion of shedding one week after vaccine administration, leading to two hypotheses: (1) that the combination of OPV and IPV shortened the duration of PV2 shedding; or (2) that IPV added to OPV suppressed the development of mucosal immunity. Exposure to environmental PV2 might have also played a role. In either case, this finding needs to be further explored.

There were some limitations of the study. tOPV continued to be used for routine and supplementary immunizations in Pakistan

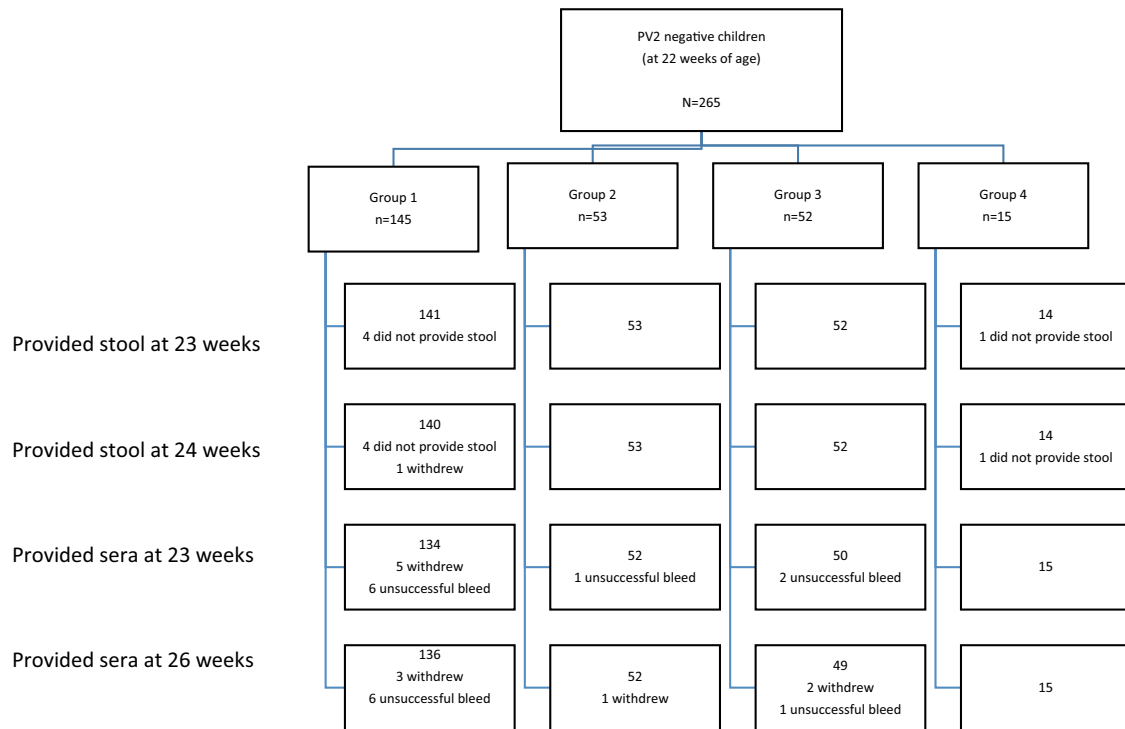


Fig. 1. Study consort chart [PV2 – Poliovirus type 2] [Group 1: tOPV, Group 2: mOPV 2, Group 3: tOPV+IPV, Group 4 mOPV 2+mIPV 2].

Table 2

Baseline demographic indicators.

Characteristic	Number/Total
Gender, Female	127/265 (48%)
Birth Weight (kg) Median (IQR)	2.85(2.53–3.12)
Monthly income of the Household (\$)	
<50	4/265 (2%)
50–100	177/265 (67%)
>100	81/265 (31%)
Exclusive breastfeeding at 22 weeks	198/265 (75%)

during the study period, resulting in potential secondary exposure to PV2 among study participants. Further, due to the delay in importation permits for mOPV2 and mIPV2, only about half of study participants that had originally been planned for received these vaccines.

Since the switch from tOPV to bOPV in April 2016, there is a growing cohort of young children that have no anti-PV2 antibodies. In addition, the immunization coverage with the newly introduced

IPV is quite low, especially in the polio high risk areas [14]. Therefore, the spread of VDPV2 from potential existing undetected foci or from a post-switch use of mOPV2 vaccines is a growing concern.

Assessment of tools to control VDPV2 outbreaks demonstrated that mOPV2 is a superior vaccine when compared to tOPV and that adding IPV to OPV significantly improves the immunological response in several ways: it improves seroconversion, increases titer of anti-PV2 antibodies, leads to faster secondary immunological response, and shortens the duration of PV2 excretion in stool.

We believe that the use of mOPV2 for control of cVDPV2 outbreaks is the correct strategy – mOPV2 is the only vaccine capable of inducing mucosal immunity in naive children. However, the use of mOPV2 has its risks, namely it may seed new VDPV2 outbreaks. Therefore the use of mOPV2 must be considered carefully; at present only the Director General of WHO may authorize the use of mOPV2.

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Table 3

Seroconversion to poliovirus type 2 among seronegative children measured one and four weeks after administration of study vaccines, median of titer calculated among seroconverted children [median titer and CI 95% calculated among seroconverted children].

		Group 1 tOPV	Group 2 mOPV2	Group 3 tOPV+IPV	Group 4 mOPV2+mIPV2
Seroconversion one week after vaccine administration	n/N (%)	38/133 (29%)	31/51 (61%)	47/47 (100%)	15/15 (100%)
	95% CI	21–37	46–74	n/a	n/a
	Median titer (CI95%)	51 (18, 178)	144 (51, 408)	576 (370, 897)	≥1448 (1167, ≥1448)
Seroconversion four weeks after vaccine administration	n/N (%)	64/133 (48%)	39/51 (76%)	46/47 (98%)	15/15 (100%)
	95% CI	39–57	63–87	89–100	n/a
	Median Titer (CI95%)	574 (221, ≥1448)	1152 (810, ≥1448)	574 (320, 1038)	≥1448 (1302, ≥1448)

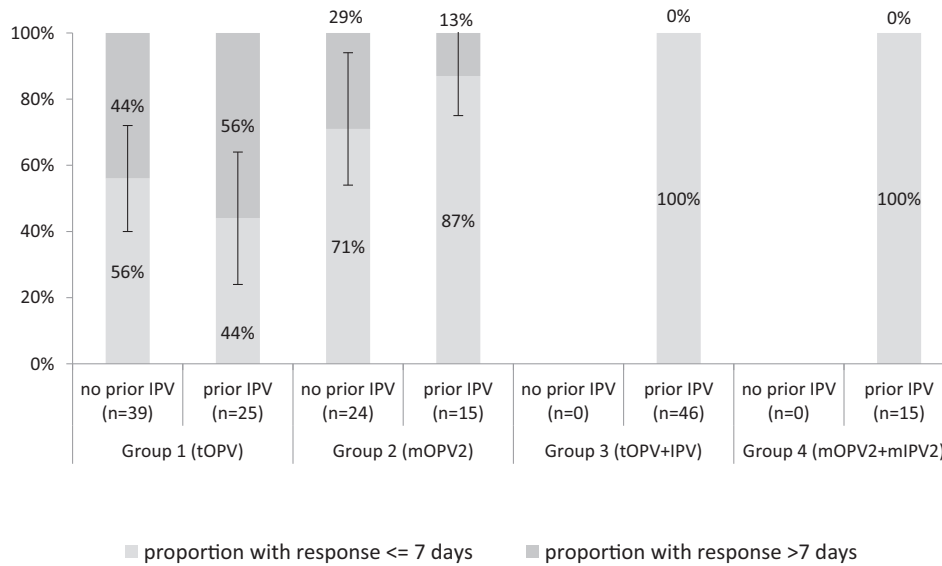


Fig. 2. Proportion of subjects who mounted an anamnestic response to poliovirus vaccine in PV2-seronegative children (measured as proportion of seroconvertors who responded to PV2 one week after vaccine administration and stratified by whether they had received prior IPV or not).

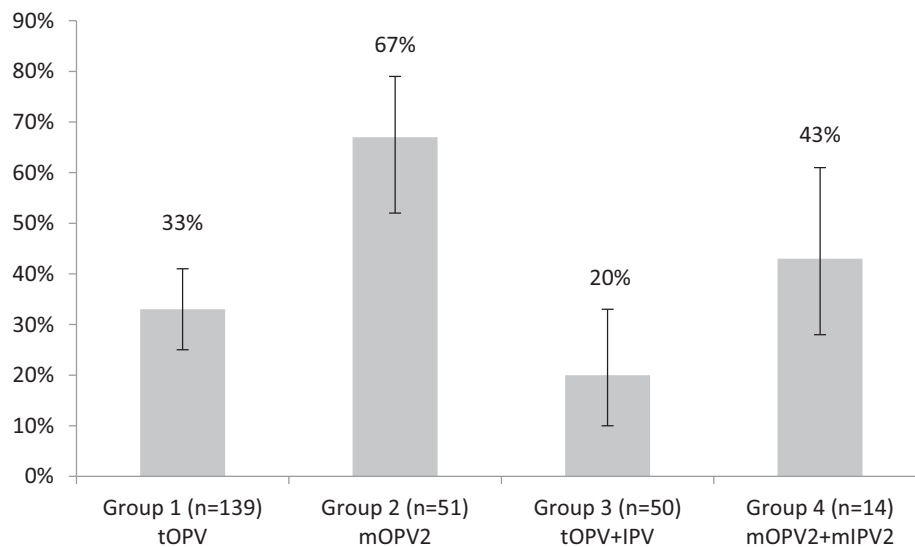


Fig. 3. Excretion of poliovirus type 2 in stool one week after OPV administration among seronegative children who were not excreting poliovirus type 2 at 22 weeks.

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Conflict of interest

All authors – no conflict of interest declared.

Disclaimer

The findings and conclusions in this report are those of the author(s) and do not necessarily represent the views of CDC and other contributing agencies.

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