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# Poliovirus type 1 systemic humoral and intestinal mucosal immunity induced by monovalent oral poliovirus vaccine, fractional inactivated poliovirus vaccine, and bivalent oral poliovirus vaccine: A randomized controlled trial

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## Abstract

**Background:** To inform response strategies, we examined type 1 humoral and intestinal immunity induced by 1) one fractional inactivated poliovirus vaccine (fIPV) dose given with monovalent oral poliovirus vaccine (mOPV1), and 2) mOPV1 versus bivalent OPV (bOPV).

**Methods:** We conducted a randomized, controlled, open-label trial in Dhaka, Bangladesh. Healthy infants aged 5 weeks were block randomized to one of four arms: mOPV1 at age 6–10–14 weeks/fIPV at 6 weeks (A); mOPV1 at 6–10–14 weeks/fIPV at 10 weeks (B); mOPV1 at 6–10–14 weeks (C); and bOPV at 6–10–14 weeks (D). Immune response at 10 weeks and cumulative response at 14 weeks was assessed among the modified intention-to-treat population, defined as seroconversion from seronegative (<1:8 titers) to seropositive (≥1:8) or a four-fold titer rise among seropositive participants sustained to age 18 weeks. We examined virus shedding after two doses of mOPV1 with and without fIPV, and after the first mOPV1 or bOPV dose. The trial is registered at ClinicalTrials.gov (NCT03722004).

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Contributors

All authors attest they meet the ICMJE criteria for authorship. All authors contributed to the design of the study. Study implementation was managed in Bangladesh by KZ, ABA, MY, WH and supported by all authors. KAVJ, LW, JLKA, BAM contributed to laboratory testing. CJS, ALW, CFE, AA, contributed to data analysis. All authors contributed to interpretation of study results, manuscript writing, and agreed with the decision to submit the manuscript.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Appendix A. Supplementary material

Supplementary data to this article can be found online at https://doi.org/10.1016/j.vaccine.2023.08.055.

**Findings:** During 18 December 2018 – 23 November 2019, 1,192 infants were enrolled (arms A:301; B:295; C:298; D:298). Immune responses at 14 weeks did not differ after two mOPV1 doses alone (94% [95% CI: 91–97%]) versus two mOPV1 doses with fIPV at 6 weeks (96% [93–98%]) or 10 weeks (96% [93–98%]). Participants who received mOPV1 and fIPV at 10 weeks had significantly lower shedding ($p < 0{\cdot}001$) one- and two-weeks later compared with mOPV1 alone. Response to one mOPV1 dose was significantly higher than one bOPV dose (79% versus 67%; $p < 0{\cdot}001$) and shedding two-weeks later was significantly higher after mOPV1 (76% versus 56%; $p < 0{\cdot}001$) indicating improved vaccine replication. Ninety-nine adverse events were reported, 29 serious including two deaths; none were attributed to study vaccines.

**Interpretation:** Given with the second mOPV1 dose, fIPV improved intestinal immunity but not humoral immunity. One mOPV1 dose induced higher humoral and intestinal immunity than bOPV.

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### Keywords

mOPV1; bOPV; fIPV; Humoral immunity; Intestinal immunity; Bangladesh

## 1. Introduction

A critical step to achieve a polio-free world is ultimate cessation of the live-attenuated bivalent oral poliovirus vaccine (bOPV, types 1 and 3) in essential childhood immunization programs worldwide, currently planned for one year after the global certification of wild poliovirus type 1 (WPV1) eradication [1–3]. OPVs induce both systemic humoral immunity that prevents paralysis and intestinal mucosal immunity that minimizes virus shedding which is key in areas where fecal-oral transmission predominates. In low OPV coverage areas, prolonged secondary vaccine virus transmission can lead to reversion of genomic attenuation sites and the emergence of vaccine-derived poliovirus (VDPV). VDPVs with evidence of transmission are called circulating VDPVs (cVDPVs) and are comparable in neuropathogenicity to WPV. Type 1 cVDPVs (cVDPV1) have been detected in 13 countries since 2011 due to underimmunized and unimmunized children. The risk of cVDPV1 emergence and spread could increase immediately after global bOPV cessation in areas with low immunization coverage as type 1 intestinal mucosal immunity wanes and birth cohorts with no type 1 intestinal immunity accumulate.

To mitigate seeding cVDPV outbreaks in the post-cessation era, judicious use of type-specific monovalent OPV (mOPV) will be necessary for outbreak responses. Since the 2016 global withdrawal of type 2 from OPV in essential immunization programs (i.e., trivalent OPV (tOPV; types 1, 2, 3) replaced with bOPV, “global switch”), the Global Polio Eradication Initiative (GPEI)’s cVDPV2 outbreak response guidelines have recommended at least two high quality vaccination campaigns [4] with monovalent OPV2 (mOPV2) due to its high immunogenicity [5], or the more genetically stable novel OPV2 (nOPV2). Extrapolating current cVDPV2 outbreak practices for cVDPV1 may be inappropriate as data on humoral and intestinal immunity from mOPV1 are limited. Previous studies had small sample sizes and different dosages from the current mOPV1, limiting the ability to draw inferences [6]. Recent studies using a dose of $\geq 10^6$ median cell culture infectious dose

(CCID50) reported immune responses in 81–90% after two doses and up to 99% after three doses; none examined intestinal mucosal immunity after 2–3 doses [7–10].

In addition to bOPV, inactivated poliovirus vaccine (IPV; types 1, 2, and 3) was introduced in essential immunization programs in all OPV-using countries in the mid-to-late 2010s to provide some level of protection against paralysis for all serotypes following the 2016 global switch [11]. For outbreak responses, the Strategic Advisory Group of Experts on Immunization (SAGE) currently recommends IPV use for specific epidemiological situations in IPV-only countries but not in countries that include OPV in essential immunization programs [1,12]. Previous studies of a supplemental IPV dose have not consistently observed a beneficial effect on humoral and intestinal immunity in OPV-vaccinated children [5,13–17]. Furthermore, IPV induces minimal intestinal mucosal immunity among OPV-na'ïve individuals [18]. Although a review of IPV use in cVDPV2 outbreaks noted the limited benefit of IPV and highlighted operational challenges [19], a few countries have opted to include IPV or fractional IPV (fIPV; 1/5 dose of IPV administered intradermally) use in response efforts. The use of fIPV in campaigns could offer cost savings and extend global IPV supplies [20,21] despite lower immunogenicity than full dose IPV. When combined with the first or second mOPV1 dose, fIPV could close the type 1 immunity gap in children who did not respond to mOPV1. No studies have examined this to our knowledge.

Independently, there is the possibility to remove type 3 from OPV and shift from bOPV to mOPV1 in vaccination campaigns and essential immunization programs with WPV type 3 declared eradicated. Yet data on the immunogenicity of mOPV1 compared with bOPV, particularly 1–2 doses, has been variable. The high variability reported for a single mOPV1 or bOPV dose for type 1 immune response (10–76%) and vaccine virus shedding upon challenge (26–76%) may be due to the first dose given at birth or administration to previously tOPV-vaccinated children [8,10,22,23].

To guide future mOPV1 and fIPV recommendations for cVDPV1 outbreak response efforts, and better understand immunogenicity of mOPV1 in relation to bOPV, we investigated type 1 systemic humoral and intestinal mucosal immunity among OPV-naïve infants induced by mOPV1 with or without fIPV, and compared with bOPV.

## 2. Methods

### 2.1. Study design and participants

We conducted a randomized, controlled, open-label phase four trial at two clinics (Mirpur and Mohakhali) in urban Dhaka, Bangladesh. Our study was designed to inform outbreak response vaccination activities and enrolling immunologically naive infants allowed us to calculate the most conservative estimates of humoral and intestinal immunity. We used 4-week intervals between vaccinations to replicate the standard 4-week intervals between vaccination campaigns. Study protocol and amendments were approved by icddr,b's Institutional Review Board and US Centers Disease Control and Prevention (CDC); CDC staff had no interaction with participants nor access to personally identifiable information. This publication adheres to CONSORT reporting guidelines [24].

Within assigned communities, fieldworkers identified expectant mothers and arranged clinic visits for interested parents. Full term (>37 weeks), healthy infants aged 5 weeks (35–41 days) from singleton births were eligible if families remained in the area for the 14-week study duration. Infants were ineligible if they had acute illness (e.g., required hospital admission, or illness prevented 6 week-visit activities) or chronic illness (e.g., immunodeficiency, blood disorder); previously poliovirus vaccinated (bOPV birth doses are not administered); or known allergies/sensitivities to poliovirus vaccines or its contents. Medical officers obtained written informed consent from parents. Participants were discontinued if parents withdrew consent; blood sample was not collected at the 6-week visit; poliovirus vaccination was received outside the study; participant started immunosuppressive medication; or developed a medical condition that posed a risk to the participant (e.g., immunodeficiency, allergy to poliovirus vaccine component).

## 2.2. Randomization and masking

Participants were randomized (1:1:1:1) to one of four study arms using varying block sizes of four and eight. Arm A received three doses of mOPV1 at 6–10–14 weeks and fIPV at 6 weeks (mOPV1 + fIPV6); arm B received three doses of mOPV1 at 6–10–14 weeks and fIPV at 10 weeks (mOPV1 + fIPV10); arm C received three doses of mOPV1 at 6–10–14 weeks (mOPV1); and arm D received three doses of bOPV at 6–10–14 weeks (bOPV). Medical officers at each clinic conducted randomization using REDCap (hosted by Vanderbilt University) [25,26]. Study staff and participants were masked until study arm assignment was revealed; only laboratory staff remained masked during and after the trial.

## 2.3. Procedures

After enrollment, follow-up activities consisted of clinic and home visits. During the 6-week clinic visit, staff obtained participant's clinical history (i.e., breastfeeding, vaccination, health status), conducted a physical examination (temperature, weight, length), collected a blood sample, administered study vaccine(s), and monitored for 30 min for any immediate systemic or injection-site adverse events. Weight (precision 100 g) and length (precision 1 mm) was measured twice; the mean was used to assess for evidence of wasting (reduced weight for age) or stunting (reduced length for age) according to WHO's Multicenter Growth Reference Study's child-growth standard curves [27]. Measurements more than two SD below the mean of the reference population indicated wasting or stunting. Participants returned for clinic visits at 10–14–18 weeks of age to complete the same activities as the 6-week visit. Fieldworkers conducted home visits to collect stool specimens at 6–7–8 weeks for participants in arms B, C, and D, and at 14–15–16 weeks for participants in arms A, B, and C. Stool collection kits were delivered one day prior to the scheduled collection date. Mothers collected and placed the first stool specimen in the container, stored in a cool place in the home, and notified fieldworkers immediately. Fieldworkers retrieved specimens within two hours of collection and placed in a study refrigerator at 2–8 °C within 30 min of pick-up. All study data were collected and managed in REDCap.

mOPV1 used in the trial was in a 20-dose vial manufactured by PT Bio Farma; each 0·1 mL dose (2 drops) contained ≥ $10^6$ $CCID_{50}$ of Sabin serotype 1. bOPV was in 20-dose vials manufactured by Sanofi Pasteur and each 0-1mL dose (2 drops) contained ≥ $10^6$ $CCID_{50}$

of Sabin serotype 1 and ≥ $10^{5.8}$ CCID50 of Sabin serotype 3. IPV was in 10-dose vials manufactured by Sanofi Pasteur and contained serotype 1 (40 D-antigen units of Mahoney strain), serotype 2 (8 D-antigen units of MEF1 strain) and serotype 3 (32 D-antigen units of Saukett strain). The fIPV dose (0–1 mL) was administered intradermally using needle & syringe on the upper right arm. Essential immunizations (except poliovirus vaccines) were also administered during eligible clinic visits according to the Expanded Programme on Immunization (EPI) of the Bangladesh Ministry of Health and Family Welfare. These were the pentavalent vaccine (diphtheria, tetanus, pertussis, *Haemophilus influenzae* type b, and hepatitis B) and pneumococcal conjugate vaccine. Upon completion of study activities, participants received catch-up bOPV and fIPV in accordance with Bangladesh's EPI recommendations. All vaccines were kept in cold chain according to manufacturer's recommendations; multi-dose vials were used in accordance with WHO's open vial policy [28].

Blood samples (1 mL) were collected by venipuncture from all participants at 6–10–14–18 weeks of age, prior to study vaccination. Stool samples (≥8 g) were collected from participants as described above. Blood and stool specimens were transported to icddr,b's laboratory at 2–8 °C by end of day. Blood samples were centrifuged within 24 h of collection and serum specimens aliquoted separately for testing (−20 °C) and reserve storage (−70 °C). Stool specimens were divided for testing and reserve storage and maintained at −20 °C. Upon completion of all study activities, serum and stool samples were shipped to the CDC laboratory in Atlanta, GA, USA, for testing. A poliovirus micro-neutralization assay was used to measure antibody titers to poliovirus serotypes 1, 2, and 3; upper limit of reciprocal antibody titer detection was ≥ 1448 [29]. Direct molecular detection via polymerase chain reaction (PCR) was used to identify vaccine virus in stools, providing a poliovirus type-specific qualitative result [30,31].

### 2.4. Outcomes

Co-primary outcomes were poliovirus type 1 immune response at 10 weeks of age and cumulative immune response at 14 weeks of age; cumulative immune response at 18 weeks was also measured as a secondary outcome. Immune response was defined as seroconversion from seronegative (<1:8 titers) to seropositive (≥1:8), or a four-fold rise in titers among seropositive participants, adjusting for the exponential decay in maternal antibodies assuming a half-life of 28 days. In addition, seropositivity (i.e., ≥1:8) had to be sustained through 18 weeks of age. Cumulative immune response was defined as immune response at any point up to and including the time of final assessment. Immune response at 10 weeks was used to evaluate differences in immunogenicity of one mOPV1 dose in comparison with one bOPV dose (arms B/C vs D); and one mOPV1 dose in comparison with the addition of fIPV at 6 weeks (arms B/C vs A). Cumulative immune response at 14 weeks was used to assess any differences in immunogenicity of two mOPV1 doses with or without fIPV at 6 weeks (arm A vs C); with or without fIPV at 10 weeks (arm B vs C); or compared with two bOPV doses (arm C vs D); a comparison was also made between two mOPV1 doses with fIPV at 6 or at 10 weeks (arm A vs B). Cumulative immune response at 18 weeks was used to identify any differences in immunogenicity of three mOPV1 doses in comparison with the addition of fIPV at 6 weeks (arm C vs A); the addition of fIPV at

10 weeks (arm C vs B); and compared with three bOPV doses (arm C vs D). Differences in immunogenicity within arm C participants from one to two mOPV1 doses (10 and 14 weeks) and two to three mOPV1 doses (14 and 18 weeks) were also evaluated.

Secondary outcomes were median reciprocal antibody titers and type 1 vaccine virus detected in stools. Median reciprocal antibody titers were calculated at the same time points as immune response (10 weeks) and cumulative immune response (14 and 18 weeks). Direct testing of intestinal immunity is challenging and was measured indirectly by the presence or absence of type 1 vaccine virus in stools ("shedding") following a first or challenge dose. Virus shedding after the first OPV dose is associated with the development of intestinal mucosal immunity ("vaccine take") whereas shedding after a subsequent OPV dose (e.g., challenge dose) suggests lack of intestinal mucosal immunity [32]. We collected stool specimens one- and two-weeks after the first mOPV1 and bOPV dose (Arms B, C, D) to measure vaccine take and after the 14-week mOPV1 challenge dose (Arms A, B, C) to measure intestinal immunity.

Systemic and injection-site adverse events were monitored during the study. Adverse events (AE) were defined as any illness during the study period and classified as serious AE (SAE) if a life-threatening event, hospitalization, paralysis or severe disability/incapacity, postvaccination anaphylaxis, or death. At each clinic visit, parents were asked of any illness since the previous visit and participants were monitored for 30 min after study vaccination. Parents were instructed to seek medical care for the participant if illness occurred between clinic visits and notify study staff as soon as feasible. The principal investigator reviewed all adverse event reports, and all serious adverse events were reported within 24 h to icddr,b's IRB, the Data Safety Monitoring Board, and CDC.

### 2.5. Statistical analysis

The sample size for the study was calculated to address the primary objectives. With a fixed enrollment target of 314 per arm (overall 1,256) and 5% attrition, we assumed 95% of participants would be seronegative or have declining maternal antibody titers at 6 weeks of age giving a sample size of 278 infants per arm. We conservatively estimated that 80% of infants would have a type 1 immune response after two doses of mOPV1 and 90% after two doses of mOPV1 and fIPV [7]. A minimum of 247 infants per arm was needed at 85% power and a two-sided μ of 0·05. A direct estimate of type 1 immune response to one bOPV dose at 6 weeks of age was unavailable in the literature, therefore we extrapolated findings from a study [23] and estimated a 70% immune response to bOPV. The unadjusted sample size of 278 in arm D (bOPV) and the combined 576 infants in arms B/C (mOPV1) at 6 weeks was sufficient to obtain 85% power with a one-sided μ of 0·05 to detect a statistically significant difference of 8.8%. A one-sided μ was used because mOPV1 immune response was assumed greater than bOPV.

Fisher's exact test was used to assess differences in type 1 immune responses, cumulative immune responses, and detection of type 1 vaccine virus in stools between study arms, and the Wilson's score method for 95% confidence intervals. The Kruskal-Wallis test was used to measure differences in reciprocal antibody titer distributions between study arms. A two-sided McNemar's test was used to assess differences in immune responses and

cumulative immune responses within participants. The Wilcoxon signed rank test with continuity correction was used to assess differences in reciprocal titer distributions within participants. Multiple comparison correction was not conducted because apriori hypotheses were examined at different end points.

Reverse cumulative distribution function curves were used to visualize differences in reciprocal antibody titers among responders with the y-axis denoting the proportion of participants with reciprocal antibody titers at the corresponding x-axis and greater. Descriptive analyses (percentages and medians) were performed for baseline characteristics, adverse events, fIPV injection site characteristics, virus shedding among immune responders, and influence of maternal antibodies. Poliovirus antibodies at baseline were assumed to represent maternal antibodies and reciprocal antibody titers ≥ 64 were categorized as “high” while titers < 64 were categorized as “low/undetectable”.

The primary analytical approach was modified intention-to-treat (mITT, participants vaccinated per arm assignment and type 1 poliovirus antibody titer results and type 1 vaccine virus shedding results available at specified time points) and per-protocol. Data were analyzed in R (version 4.1.3). This trial is registered at ClinicalTrials.gov (number NCT03722004).

### 2.6. Role of the funding source

The sponsor of the study participated in study design, protocol development, data analysis, data interpretation, and manuscript development. The sponsor did not participate in data collection. The corresponding, first, and senior author had full access to all study data, except personally identifiable information. All authors agreed with the decision to submit for publication.

## 3. Results

A total of 1,301 parents were approached for the study; 1,256 infants were enrolled from 18 December 2018 to 23 November 2019 (Fig. 1). The mITT population included 1,192 (95%) participants. Results are also presented for the 1,078 (86%) in the per-protocol population when findings differed (Supplemental Tables 1–3). Baseline characteristics are summarized in Table 1.

Type 1 immune response at 10 weeks among the 470 of 573 participants who received one dose of mOPV1 (arms B/C, 79% [95% confidence interval (CI): 76–82%]) was comparable to 255 of 301 participants who received mOPV1 and fIPV (arm A, 85% [80–89%]); an incidental finding was a significant difference in response between arms B and C (248 of 295 participants (84% [79–88%]) versus 222 of 298 participants (75% [69–79%]), p = 0·005) (Table 2). Median reciprocal antibody titers were also similar (arm A: ≥1448, [interquartile range (IQR): 1152-≥1448]; arms B/C: ≥1448 [IQR: ≥1448-≥1448]; p = 0.13) (Fig. 2).

Compared with two mOPV1 doses (arm C, 281 of 298 participants (94% [91–97%])), no significant differences in type 1 cumulative immune response at 14 weeks were observed

when fIPV was additionally given at 6 weeks (arm A, 289 of 301 participants (96% [93–98%])) or 10 weeks (arm B, 284 of 295 participants (96% [93–98%])) (Table 2). Median reciprocal antibody titers and IQR reached the upper limit of detection (≥1448 [IQR: ≥1448-≥1448]) for all three arms (Fig. 2); titer distributions differed (p = 0·019) when fIPV was administered at 10 weeks (arm B) in comparison with mOPV1 only (arm C). Cumulative immune response of two mOPV1 doses with fIPV at 6 weeks (arm A, 289 of 301 participants (96% [93–98%])) or fIPV at 10 weeks (arm B, 284 of 295 participants (96% [93–98%])) did not differ (Table 2). Titer distributions differed (p = 0·042) but this was not observed in the per-protocol analysis (p = 0·11) (Supplemental Table 2).

Type 1 cumulative immune response at 18 weeks did not differ for three mOPV1 doses (arm C, 295 of 298 participants (99% [97–100%])) compared with the addition of fIPV at 6 weeks (arm A, 296 of 301 participants (98% [96–99%])) or fIPV at 10 weeks (arm B, 289 of 295 participants (98% [96–99%])) (Table 2). Median reciprocal antibody titers and IQR reached the upper limit of detection (≥1448 [IQR: ≥1448-≥1448]) in all three arms (Fig. 2).

The gain in type 1 immunity after each mOPV1 dose was assessed among the 298 participants who received three doses of mOPV1 (arm C). There was a statistically significant increase (p < 0·0001) in immunity after the second mOPV1 dose (281 participants (94% [91–97%])) compared with the first dose (222 participants (75% [69–73%])) and a significant increase (p < 0·0001) after the third mOPV1 dose (295 participants (99% [97–100%]). At every dose level, the median reciprocal antibody titers and IQR reached the upper limit of detection (≥1448 [IQR: ≥1448-≥1448]). No significant difference was observed in titer distributions among responders between one and two doses (p = 0·45). Although titers among responders did not differ for two and three doses (p = 0·069), a significant difference was detected in the per-protocol analysis (p = 0·020).

Differences in type 1 immunity were observed for mOPV1 in comparison with bOPV. One mOPV1 dose (arms B/C) elicited an immune response among 470 of 593 participants (79% [76–82%]) and was significantly higher (p = 0·0002) compared with one bOPV dose (arm D) among 201 of 298 participants (67% [62–73%]) (Table 2). Differences in antibody titers among mOPV1 responders [≥1448 (IQR: ≥1448-≥1448] compared with bOPV responders [≥1448 (IQR: ≥910-≥1448] were also observed (p < 0·0001) (Fig. 2). Cumulative immune response to two doses of mOPV1 (281 of 298 participants (94% [91–97%])) was significantly higher (p = 0·026) than two doses of bOPV (265 of 298 participants (89% [85–92%])) although no significant difference was observed in the per-protocol analysis (Table 2, Supplemental Table 2).

Reciprocal antibody titers did not differ. Cumulative immune response to three mOPV1 doses (295 of 298 participants (99% [97–100%])) was higher (p = 0·033) than three bOPV doses (286 of 298 participants (96% [93–98%])); however, this was not significant in the per-protocol analysis (Supplemental Table 2). Median reciprocal antibody titers and IQR reached the upper limit of detection (≥1448 [IQR: ≥1448-≥1448]).

There was no significant difference in type 1 vaccine virus shedding one week after the first dose of mOPV1 or bOPV; 493 of 581 participants (85% [82–88%]) who received

one mOPV1 dose (arms B/C) were shedding compared with 233 of 290 participants (80% [75–85%]) who received one bOPV dose (arm D; p = 0·10) (Table 3, Fig. 3A). However, two weeks after the first dose a significantly higher percentage of participants were shedding (p < 0·0001) among those who received mOPV1 (443 of 581 participants (76% [73–80%])) compared with bOPV (162 of 290 participants (56% [50–62%])) indicating higher vaccine take among mOPV1 recipients. An incidental finding was the significant difference (p = 0·015) in virus shedding among those who received one mOPV1 dose (arms B (234 of 290 participants (81% [76–85%]) and C (209 of 291 participants (72% [66–77%]), Table 3). A higher percentage of participants who had an immune response at 10 weeks of age also had virus detected in stool specimens one- and two-weeks after the first dose compared with those who did not have an immune response at 10 weeks of age, suggesting an association between vaccine take and development of immune response (Supplemental Fig. 1).

Differences in type 1 vaccine virus shedding varied after the 14-week mOPV1 challenge dose for participants who received two doses of mOPV1 with or without fIPV (Table 3, Fig. 3B). Restricted to participants not shedding at 14 weeks, no difference was identified one- or two-weeks post challenge dose for two doses of mOPV1 with or without fIPV at 6 weeks. Vaccine virus one-week post challenge dose was detected in 52 of 244 participants (21% [17–27%]) who received two doses of mOPV1 compared with 51 of 254 participants (20% [15–26%]) who received two doses of mOPV1 and fIPV at 6 weeks. Vaccine virus two-weeks post-challenge dose was detected in 35 of 244 participants (14% [10–20%]) who received two doses of mOPV1 compared with 25 of 254 participants (10% [6–14%]) who received two doses and fIPV at 6 weeks. However, participants who received two doses of mOPV1 and fIPV at 10 weeks had significantly lower shedding one- and two-weeks post-challenge dose compared with participants who received mOPV1 only. Only 10% [6–14%] of participants (24 of 249) who received two doses of mOPV1 and fIPV at 10 weeks had virus detected one-week post challenge and 6% [3–10%] of participants (14 of 249) shed virus two-weeks post challenge dose. This was in comparison to the 21% [17–27%] one week post challenge (p = 0·0004) and 14% [10–20%] two weeks post challenge (p = 0·0014) previously reported for two mOPV1 doses alone.

There were 615 fIPV doses administered (arm A = 312, arm B = 303); median bleb size was 8 mm (range: 3–10 mm, IQR: 6–8 mm). No residual vaccine on the skin (i.e., wetness) was reported for 525 (85%) injections. Of the 58 small blebs (≤5mm), 53 (91%) had no wetness. No redness, swelling, bruising, or lacerations were observed at the injection site after 30 min.

Potential interference of maternal antibodies on type 1 immunity development could not be assessed due to the high percentage of immunity (>95%) in all arms at 18 weeks of age.

Poliovirus types 2 and 3 immune responses and median reciprocal antibody titers are summarized in Table 4.

Ninety-nine AEs were reported among 90 participants; 29 were SAEs including two deaths (Table 5). AEs (p = 0·11) and SAEs (p = 0·56) did not differ among arms. None of the AEs were attributed to the study vaccines.

## 4. Discussion

Several key findings from this study can be used to inform type 1 outbreak response efforts at present and after bOPV cessation. First, interrupting virus transmission is essential, and the slightly improved intestinal mucosal immunity observed when fIPV is given with the second mOPV1 dose could be important to interrupting transmission. A single fIPV dose appeared to boost type 1 intestinal immunity in infants who previously received one mOPV1 dose (11% difference after one week, 8% difference after two weeks), consistent with current understanding of IPV as a booster in OPV-vaccinated individuals [33]. A single fIPV dose did not affect type 1 systemic humoral immunity when given with one, two, or three doses of mOPV1. After three doses of mOPV1, cumulative immune response for all three arms was ≥98%. Our overall findings on a single fIPV dose are consistent with a recent review on IPV use in cVDPV2 outbreaks that concluded some small improvement in intestinal immunity may be gained in previously OPV-vaccinated children; we agree with review authors that the decision to use fIPV must be weighed against operational challenges [19]. If fIPV is to be used for outbreak response efforts, countries should consider use after at least one mOPV1 campaign to maximize its benefit on intestinal mucosal immunity.

Our findings indicate three doses of mOPV1 may be needed to achieve high levels of systemic humoral and intestinal mucosal immunity. We found type 1 systemic humoral immunity reached 94% with two doses and 99% with three doses, consistent with most other studies that examined two (81–90%) and three (99%) doses of mOPV1 [8–10,22]. However in a field setting, vaccine take is likely to be lower than a clinical trial setting and vaccines may not reach >90% of target populations [4]. To reach high levels of type 1 immunity, at least three high quality mOPV1 vaccination campaigns should be considered.

We also found one dose of mOPV1 to be more immunogenic than bOPV but this difference diminished with subsequent doses. There was a 13% difference in immune response after one dose of mOPV1 compared with bOPV (79% v 67%; $p = 0{\cdot}0002$) and antibody titer distributions differed ($p < 0{\cdot}0001$). This gap declined to 5% after the second dose (94% versus 89%; $p = 0{\cdot}026$) and 3% after the third dose (99% versus 96%; $p = 0{\cdot}033$). While the two- and three-dose results were not significantly different in our per-protocol analyses, the narrowing gap in humoral immunity remains important in preventing paralysis. We also observed differences in mOPV1 and bOPV in intestinal mucosal immunity development. Vaccine virus shedding two weeks after the first dose was 20% higher in the mOPV1 arm (76% v 56%; $p < 0{\cdot}0001$) suggesting better vaccine take than bOPV. The observation between vaccine take and immune response at 10 weeks also suggests that a single mOPV1 dose was better at inducing vaccine replication (and therefore intestinal mucosal immunity) than a single bOPV dose even though intestinal immunity was not measured after a challenge dose. While differences in per dose take and immunogenicity are expected due to type 1 and 3 interference in intestinal replication for bOPV, our findings suggest at least three high quality rounds of bOPV are necessary to achieve ≥ 90% response to type 1 poliovirus. Faced with similar challenges as previously described for mOPV1 campaigns, it may be prudent to conduct at least four bOPV campaigns to achieve high levels of immunity in a field setting and reach targeted children.

There are limitations to our study. We selected polio-vaccine naïve young infants to minimize interference from essential immunizations and background community exposure. However, maternal antibodies and the immature immune system of participants may have hampered the effectiveness of fIPV compared with older children. However, mOPV1 or bOPV responses may be higher in our participants than older children living in areas where malnutrition and diarrheal diseases are highly prevalent. Despite these limitations, we believe our findings are conservative estimates of the number of vaccinations needed and are generalizable to areas with little to no poliovirus immunity as expected in cVDPV outbreak-affected areas and post-bOPV cessation. Although we conducted a randomized controlled trial and group assignment was blinded, we cannot explain the significant difference in arms B versus C after a single mOPV1 dose for immune response (84% v 75%; p = 0·005) or virus shedding two weeks thereafter (81% v 72%; p = 0·015). No differences in enrollment, clinic site, inadvertent fIPV administration, baseline characteristics, or secondary type 1 exposure were noted. The combined arms B/C results were presented as a conservative estimate that did not affect interpretation of results. Selection bias is likely minimal as 94–96% of enrolled participants in each arm were included in the mITT population. A few findings from mITT analyses differed from the per-protocol analysis due to ~ 10% drop in participants across all arms, particularly our mOPV1 and bOPV response comparisons. Secondary exposure to OPV1 from the community may have occurred due to bOPV use in Bangladesh's essential immunization program. Finally, we extrapolated our mOPV1 challenge findings to indicate protection against WPV1 and cVDPV1.

Current GPEI plans are to use bOPV to respond to type 1 poliovirus outbreaks (cVDPV1 and WPV1) until bOPV cessation in essential immunization programs after which mOPV1 will be used. Our study suggests that in an extreme setting of sizable OPV1-naïve children, two vaccination campaigns is insufficient to achieve ≥ 90% response to type 1 when bOPV is used, and unlikely when mOPV1 is used. Rather, the minimally recommended number of vaccination campaigns may need to be vaccine specific. Vaccine choice may also need to be guided by the epidemiology of the outbreak – consider bOPV for co-circulation of types 1 and 3 and mOPV1 for type 1 outbreaks in inaccessible areas to quickly achieve high immunity levels. Essential to successfully interrupting transmission is careful planning to overcome operational limitations in reaching all targeted children. Phase 1 trials are underway for more attenuated Sabin strains for types 1 (nOPV1) and 3 (nOPV3) that were designed to further reduce the risk of reversion and are likely to become the vaccine(s) of choice for type-specific poliovirus outbreaks. Our study adds to the growing body of literature on fIPV use in outbreak response efforts; while our findings neither contradict nor support SAGE's recommendation of not using fIPV in OPV-using countries, they highlight that the decision should carefully consider epidemiological context.

## Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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### Disclaimer

The findings and conclusions in this report are those of the authors and do not necessarily represent the official position of the U.S. Centers for Disease Control and Prevention.

## Data Sharing

The study is registered on the clinicaltrials.gov website (NCT03722004), aggregated data from Tables 2–3 will be added to the registration with publication. In accordance with the protocol, icddr,b investigators will have access to participant data with identifiers; external investigators will have access to de-identified participant data; de-identified data may be shared with national and international vaccine manufacturers and regulatory authorities upon request; and no participant-level data will be shared further.

## Data availability

The authors do not have permission to share data.

## References

[1]. Meeting of the Strategic Advisory Group of Experts on Immunization, April 2022: conclusions and recommendations. 2022. https://www.who.int/publications/i/item/who-wer9724-261-276 (accessed 7 Jul 2022).

[2]. Aylward RB, Sutter RW, Policy HDL. OPV cessation-the final step to a “polio-free” world. Science 2005;310(5748):625–6. [PubMed: 16254173]

[3]. Global Polio Eradication Initiative. Polio Eradication Strategy 2022-2026: Delivering on a Promise. 2021. https://polioeradication.org/gpei-strategy-2022-2026/(accessed 19 Oct 2022).

[4]. Zaman K, Estivariz CF, Morales M, et al. Immunogenicity of type 2 monovalent oral and inactivated poliovirus vaccines for type 2 poliovirus outbreak response: an open-label, randomised controlled trial. Lancet Infect Dis 2018;18(6):657–65. [PubMed: 29571817]

[5]. Global Polio Eradication Initiative. Standard Operating Procedures: Responding to a poliovirus event or outbreak 2022. https://polioeradication.org/wp-content/uploads/2022/07/Standard-Operating-Procedures-For-Responding-to-a-Poliovirus-Event-Or-Outbreak-20220807-EN-Final.pdf (accessed 26 Apr 2022).

[6]. Caceres VM, Sutter RW. Sabin monovalent oral polio vaccines: review of past experiences and their potential use after polio eradication. Clin Infect Dis 2001;33 (4):531–41. [PubMed: 11462191]

[7]. Mir F, Quadri F, Mach O, et al. Monovalent type-1 oral poliovirus vaccine given at short intervals in Pakistan: a randomised controlled, four-arm, open-label, non-inferiority trial. Lancet Infect Dis 2015;15(8):889–97. [PubMed: 26093979]

[8]. el-Sayed N, el-Gamal Y, Abbassy AA, et al. Monovalent type 1 oral poliovirus vaccine in newborns. N Engl J Med 2008;359(16):1655–65. [PubMed: 18923170]

[9]. Estivariz CF, Anand A, Gary HE Jr, et al. Immunogenicity of three doses of bivalent, trivalent, or type 1 monovalent oral poliovirus vaccines with a 2 week interval between doses in Bangladesh: an open-label, non-inferiority, randomised, controlled trial. Lancet Infect Dis 2015;15(8):898–904. [PubMed: 26093980]

[10]. Sutter RW, John TJ, Jain H, et al. Immunogenicity of bivalent types 1 and 3 oral poliovirus vaccine: a randomised, double-blind, controlled trial. Lancet 2010;376 (9753):1682–8. [PubMed: 20980048]

[11]. Meeting of the Strategic Advisory Group of Experts on immunization, April 2013 - conclusions and recommendations, https://www.ncbi.nlm.nih.gov/pubmed/23696983 (accessed 22 Sep 2018).

[12]. Strategic Advisory Group of Experts on Immunization (SAGE). SAGE October 2022 Meeting Highlights: Strategic Advisory Group of Experts on Immunization (SAGE), 2022. https://cdn.who.int/media/docs/default-source/immunization/sage/2022/october/highlights_sage_oct_2022.pdf?sfvrsn=69f947c4_5 (accessed 18 Oct 2022).

[13]. Jafari H, Deshpande JM, Sutter RW, et al. Polio eradication. Efficacy of inactivated poliovirus vaccine in India. Science 2014;345(6199):922–5. [PubMed: 25146288]

[14]. John J, Giri S, Karthikeyan AS, et al. Effect of a single inactivated poliovirus vaccine dose on intestinal immunity against poliovirus in children previously given oral vaccine: an open-label, randomised controlled trial. Lancet 2014;384(9953): 1505–12. [PubMed: 25018120]

[15]. Moriniere BJ, van Loon FP, Rhodes PH, et al. Immunogenicity of a supplemental dose of oral versus inactivated poliovirus vaccine. Lancet 1993;341(8860): 1545–50. [PubMed: 8099637]

[16]. Parent du Chatelet I, Merchant AT, Fisher-Hoch S, et al. Serological response and poliovirus excretion following different combined oral and inactivated poliovirus vaccines immunization schedules. Vaccine 2003;21(15): 1710–8. [PubMed: 12639494]

[17]. Sutter RW, Suleiman AJ, Malankar P, et al. Trial of a supplemental dose of four poliovirus vaccines. N Engl J Med 2000;343(ll):767–73. [PubMed: 10984564]

[18]. Hird TR, Grassly NC. Systematic review of mucosal immunity induced by oral and inactivated poliovirus vaccines against virus shedding following oral poliovirus challenge. PLoS Pathog 2012;8(4):e1002599. [PubMed: 22532797]

[19]. Estivariz CF, Kovacs SD, Mach O. Review of use of inactivated poliovirus vaccine in campaigns to control type 2 circulating vaccine derived poliovirus (cVDPV) outbreaks. Vaccine 2022.

[20]. Bahl S, Verma H, Bhatnagar P, et al. Fractional-dose inactivated poliovirus vaccine immunization campaign - Telangana State, India, June 2016. MMWR Morb Mortal Wkly Rep 2016;65(33):859–63. [PubMed: 27559683]

[21]. Estivariz CF, Pallansch MA, Anand A, et al. Poliovirus vaccination options for achieving eradication and securing the endgame. Curr Opin Virol 2013;3(3): 309–15. [PubMed: 23759252]

[22]. John TJ, Jain H, Ravishankar K, et al. Monovalent type 1 oral poliovirus vaccine among infants in India: report of two randomized double-blind controlled clinical trials. Vaccine 2011;29(34):5793–801. [PubMed: 21641951]

[23]. Waggie Z, Geldenhuys H, Sutter RW, et al. Randomized trial of type 1 and type 3 oral monovalent poliovirus vaccines in newborns in Africa. J Infect Dis 2012;205 (2):228–36. [PubMed: 22158680]

[24]. Schulz KF, Altman DG, Moher D, for the CONSORT Group. CONSORT 2010 Statement: updated guidelines for reporting parallel group randomised trials. https://www.goodreports.org/reporting-checklists/consort/(accessed 5 April 2023).

[25]. Harris PA, Taylor R, Thielke R, Payne J, Gonzalez N, Conde JG. Research electronic data capture (REDCap) – a metadata-driven methodology and workflow process for providing translational research informatics support. J Biomed Inform 2009;42(2): 377–81. [PubMed: 18929686]

[26]. Harris PA, Taylor R, Minor BL, Elliott V, Fernandez M, O'Neal L, McLeod L, Delacqua G, Delacqua F, Kirby J, Duda SN, REDCap Consortium. The REDCap consortium: Building an international community of software partners. J Biomed Inform 2019;95:103208. [PubMed: 31078660]

[27]. WHO. WHO child growth standards: length/height-for-age, weight-for-age, weight-for-length, weight-for-height and body mass index-for-age: methods and development. Geneva: World

Health Organization, 2006. https://www.who.int/tools/child-growth-standards/who-multicentre-growth-reference-study (accessed 17 Jul 2018).

[28]. WHO. WHO Policy Statement: Multi-dose Vial Policy (MDVP). Handling of multidose vaccine vials after opening. 2014. https://www.who.int/publications/i/item/WHO-IVB-14.07 (accessed 27 July 2022).

[29]. Weldon WC, Oberste MS, Pallansch MA. Standardized methods for detection of poliovirus antibodies. Methods Mol Biol 2016;1387:145–76. [PubMed: 26983734]

[30]. Gerloff N, Mandelbaum M, Pang H, et al. Direct detection of polioviruses using a recombinant poliovirus receptor. PLoS One 2021,16(11):e0259099. [PubMed: 34727100]

[31]. Harrington C, Sun H, Jeffries-Miles S, et al. Culture-independent detection of poliovirus in stool samples by direct RNA extraction. Microbiol Spectr 2021;9(3): e0066821. [PubMed: 34756088]

[32]. Sutter RW, Kew OM, Cochi SL, Aylward RB. Poliovirus vaccine-live. Plotkin's vaccines: Elsevier 2018::866–917. el6.

[33]. Connor RI, Brickley EB, Wieland-Alter WF, et al. Mucosal immunity to poliovirus. Mucosal Immunol 2022;15(1):1–9. [PubMed: 34239028]

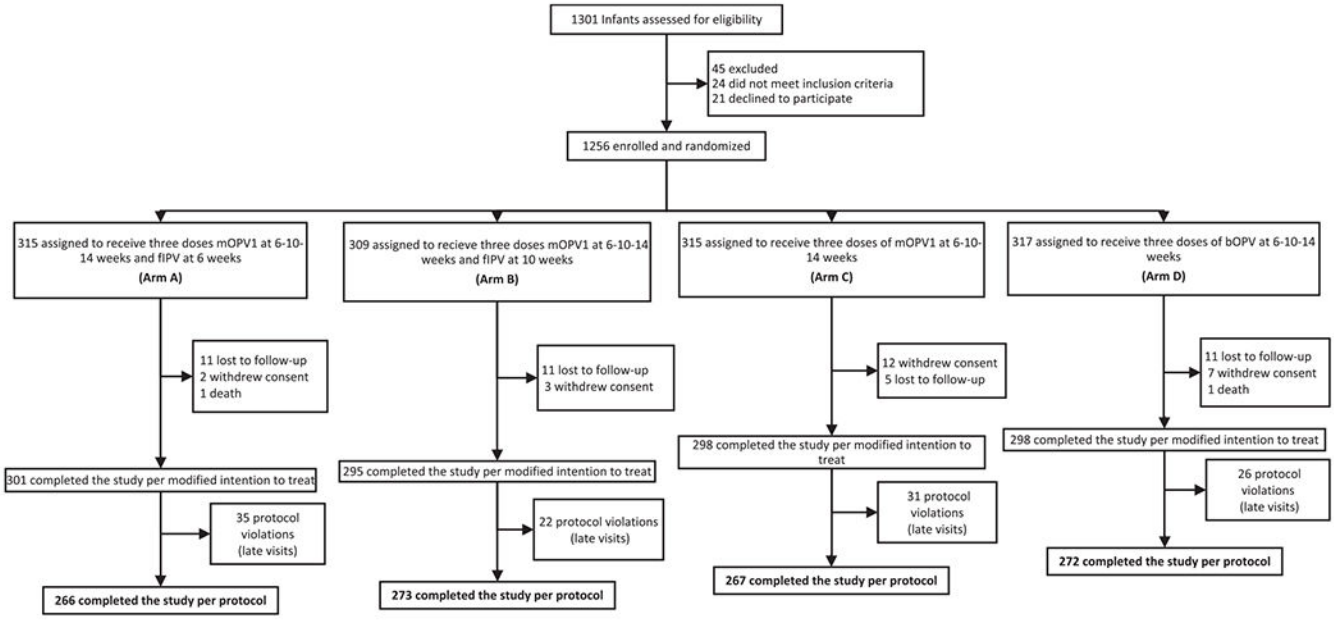


**Fig. 1. Trial profile, Bangladesh, 2018–2019.**

mOPV = monovalent oral poliovirus vaccine. fIPV = fractional inactivated poliovirus vaccine. bOPV = bivalent oral poliovirus vaccine.

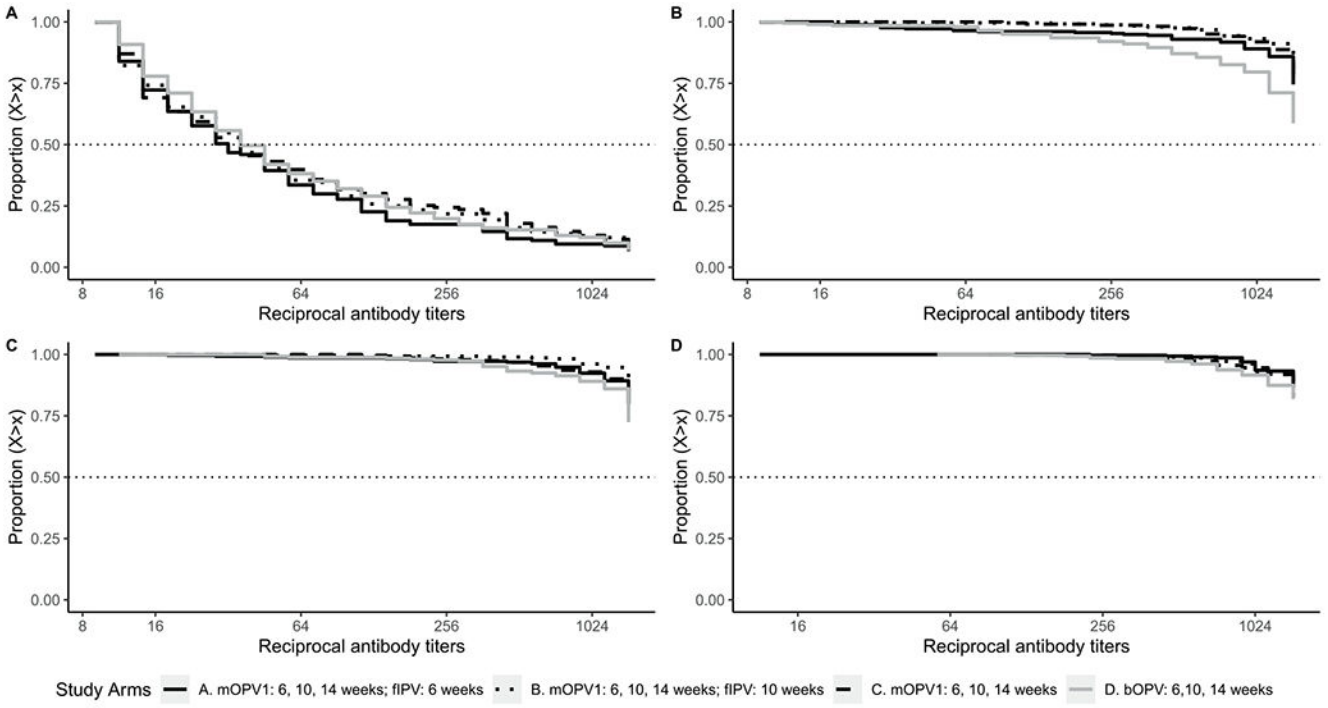


**Fig. 2. Reverse cumulative distribution function curves of reciprocal antibody titers to poliovirus type 1 by study arm**

The proportion of participants (y-axis) with measured reciprocal antibody titers and greater (x-axis) (A) among those seropositive at 6 weeks, (B) immune responders at 10 weeks, (C) cumulative immune responders at 14 weeks and (D) cumulative immune responders at 18 weeks. mOPV = monovalent oral poliovirus vaccine. fIPV = fractional inactivated poliovirus vaccine. bOPV = bivalent oral poliovirus vaccine.

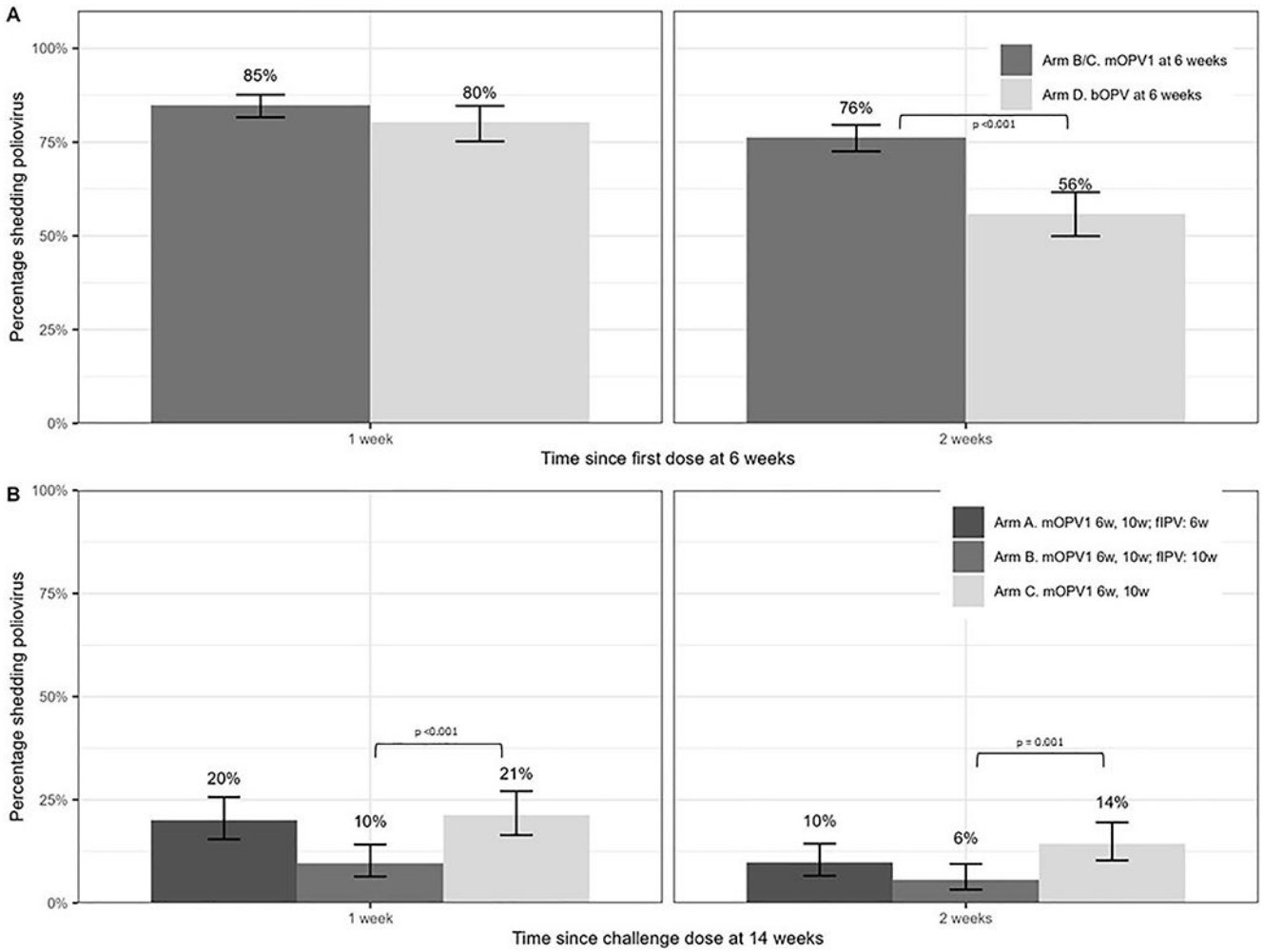


**Fig. 3. Poliovirus type 1 vaccine virus detected in stool specimens by selected study arms**

Proportion of participants (y-axis) with poliovirus type 1 vaccine virus detected from stool specimens one-week and two-weeks (A) after the first dose among participants who were not shedding at 6 weeks and (B) after a challenge dose among participants not shedding at 14 weeks. mOPV = monovalent oral poliovirus vaccine. fIPV = fractional inactivated poliovirus vaccine. bOPV = bivalent oral poliovirus vaccine, w = weeks.

**Table 1**

Baseline characteristics of the modified intention-to-treat population.

| **Baseline Characteristics** | **Arm A mOPV1 + fIPV6 (n = 301)** | | **Arm B mOPV1 + fIPV10 (n = 295)** | | **Arm C mOPV1 (n = 298)** | | **Arm D bOPV (n = 298)** | |
|---|---|---|---|---|---|---|---|---|
| Age (days) | 39 (37–40) | | 39 (37–40) | | 39 (37–41) | | 39 (37–40) | |
| Male | 148 | 49% | 161 | 55% | 143 | 48% | 160 | 54% |
| Mother's education | | | | | | | | |
| No formal school | 32 | 11% | 31 | 11% | 31 | 10% | 33 | 11% |
| Primary | 126 | 42% | 127 | 43% | 127 | 43% | 135 | 45% |
| Middle | 69 | 23% | 78 | 26% | 82 | 28% | 67 | 23% |
| High | 64 | 21% | 52 | 18% | 49 | 16% | 55 | 19% |
| Graduate | 10 | 3% | 7 | 2% | 9 | 3% | 8 | 3% |
| Exclusive breastfeeding | 57 | 19% | 67 | 23% | 57 | 19% | 68 | 23% |
| Wasting present | 13 | 4% | 20 | 7% | 15 | 5% | 11 | 4% |
| Stunting present | 34 | 11% | 31 | 11% | 34 | 11% | 43 | 14% |
| Type 1 poliovirus | | | | | | | | |
| Seropositive | 137 | 46% | 124 | 42% | 123 | 41% | 131 | 44% |
| Reciprocal antibody titers | 28.4 | (11.3–90.5) | 28.4 | (11.3–121.3) | 28.4 | (11.3–162.5) | 28.4 | (14.2–113.8) |
| Virus shedding | 0 | 0% | 4 | 1% | 6 | 2% | 6 | 2% |
| Type 2 poliovirus | | | | | | | | |
| Seropositive | 155 | 52% | 158 | 54% | 160 | 54% | 163 | 55% |
| Reciprocal antibody titers | 22.6 | (11.3–40.6) | 18.0 | (11.3–45.3) | 18.0 | (11.3–45.3) | 22.6 | (11.3–51.1) |
| Virus shedding | 0 | 0% | 0 | 0% | 0 | 0% | 0 | 0% |
| Type 3 poliovirus | | | | | | | | |
| Seropositive | 79 | 26% | 75 | 25% | 77 | 26% | 87 | 29% |
| Reciprocal antibody titers | 22.6 | (14.2–113.8) | 18.0 | (11.3–64.4) | 18.0 | (11.3–90.5) | 18.0 | (11.3–36.0) |
| Virus shedding | 0 | 0% | 7 | 2% | 7 | 2% | 11 | 4% |

Data are n (%), median (interquartile range) for age in days and reciprocal antibody titers among seropositive participants. Baseline measurements for participants were obtained at 6 weeks of age. mOPV = monovalent oral poliovirus vaccine. fIPV = fractional inactivated poliovirus vaccine. bOPV = bivalent oral poliovirus

**Table 2**

Summary of poliovirus type 1 immune response and reciprocal antibody titers by study arms.

| | Arm A mOPV1 + fIPV6 (n = 301) | | Arm B mOPV1 + fIPV10 (n = 295) | | Arm C mOPV1 (n = 298) | | Arm D bOPV (n = 298) | | Fisher's Exact Test or Kruskal-Wallis Test |
|---|---|---|---|---|---|---|---|---|---|
| One dose, 10 weeks | | | | | | | | | |
| Immune response | 255 | 85% (80–89%) | 248 | 84% (79–88%) | 222 | 75% (69–79%) | 201 | 67% (62–73%) | A vs B/C: p = 0.057<br>B/C vs D: p = 0.0002 |
| Reciprocal antibody titers | ≥1448 | (1152-≥1448) | ≥1448 | (≥1448-≥1448) | ≥1448 | (≥1448-≥1448) | ≥1448 | (910.2-≥1448) | A vs B/C: p = 0.13<br>B/C vs D: p < 0.0001 |
| Two doses, 14 weeks | | | | | | | | | |
| Cumulative immune response | 289 | 96% (93–98%) | 284 | 96% (93–98%) | 281 | 94% (91–97%) | 265 | 89% (85–92%) | A vs C: p = 0.35<br>B vs C: p = 0.33<br>A vs B: p = 1.0<br>C vs D: p = 0.026 |
| Reciprocal antibody titers | ≥1448 | (≥1448-≥1448) | ≥1448 | (≥1448-≥1448) | ≥1448 | (≥1448-≥1448) | ≥1448 | (1152-≥1448) | A vs C: p = 0.76<br>B vs C: p = 0.019<br>A vs B: p = 0.042<br>C vs D: p = 0.094 |
| Three doses, 18 weeks | | | | | | | | | |
| Cumulative immune response | 296 | 98% (96–99%) | 289 | 98% (96–99%) | 295 | 99% (97–100%) | 286 | 96% (93–98%) | A vs C: p = 0.73<br>B vs C: p = 0.34<br>C vs D: p = 0.033 |
| Reciprocal antibody titers | ≥1448 | (≥1448-≥1448) | ≥1448 | (≥1448-≥1448) | ≥1448 | (≥1448-≥1448) | ≥1448 | (≥1448-≥1448) | A vs C: p = 0.19<br>B vs C: p = 0.37<br>C vs D: p = 0.62 |

Data are the percentage of participants with immune response expressed as n/N and percentage including 95% confidence interval (CI). Immune response defined as seroconversion from seronegative (<1:8) to seropositive (≥1:8), or a four-fold rise in antibody titers among seropositives adjusted for maternal antibody decay, and seropositivity maintained through 18 weeks of age. Cumulative immune response was defined as immune response at any point up to and including at the time of assessment. Reciprocal antibody titers are presented as the median (interquartile range) among responders. mOPV1 = monovalent oral poliovirus vaccine type 1, fIPV = fractional inactivated poliovirus vaccine, bOPV = bivalent oral poliovirus vaccine. Fisher's Exact test was used to test for inequality of proportions between study arms. Kruskal-Wallis Test was used to test for inequality of antibody titer distributions between study arms.

**Table 3**

Summary of poliovirus type 1 vaccine virus detected among participants by select study arms.

| Vaccine virus shedding | Arm A mOPV1 + fIPV6 (n = 301) | | Arm B mOPV1 + fIPV10 (n = 295) | | Arm C mOPV1 (n = 298) | | Arm D bOPV (n = 298) | | Fisher's Exact Test |
|---|---|---|---|---|---|---|---|---|---|
| Baseline *, 6 weeks | – | – | 4/294 | 1% (0–4%) | 6/297 | 2% (1–5%) | 6/296 | 2% (1–5%) | |
| One week, 7 weeks | – | – | 253/290 | 87% (83–91%) | 240/291 | 83% (78–87%) | 233/290 | 80% (75–85%) | B/C v D: p = 0.10 |
| Two weeks, 8 weeks | – | – | 234/290 | 81% (76–85%) | 209/291 | 72% (66–77%) | 162/290 | 56% (50–62%) | B/C v D: p < 0.0001 |
| Baseline *, 14 weeks | 41/295 | 14% (10–19%) | 39/288 | 14% (10–18%) | 50/294 | 17% (13–22%) | – | – | |
| One week, 15 weeks | 51/254 | 20% (15–26%) | 24/249 | 10% (6–14%) | 52/244 | 21% (17–27%) | | | A vs C: p = 0.74<br>B vs C: p = 0.0004 |
| Two weeks, 16 weeks | 25/254 | 10% (6–14%) | 14/249 | 6% (3–10%) | 35/244 | 14% (10–20%) | | | A v C: p = 0.13<br>B vs C: p = 0.0014 |

Data are the percentage of participants with vaccine virus detected in stool specimens expressed as n/N and percentage including 95% confidence interval (CI).

* Restricted to participants who were not shedding at baseline (either 6 weeks or 14 weeks depending on objective) and had stool specimen results available one week and two weeks post-vaccination. mOPV1 = monovalent oral poliovirus vaccine type 1, fIPV = fractional inactivated poliovirus vaccine, bOPV = bivalent oral poliovirus vaccine. Fisher's Exact test was used to test for inequality of proportions between study arms.

**Table 4**

Summary of poliovirus types 2 and 3 immune response and reciprocal antibody titers by study arms, modified intention-to-treat population.

| | Arm A mOPV1 + fIPV6 (n = 301) | | Arm B mOPV1 + fIPVIO (n = 295) | | Arm C mOPV1 (n = 298) | | Arm D bOPV (n = 298) | |
|---|---|---|---|---|---|---|---|---|
| **Type 2** | | | | | | | | |
| One dose, 10 weeks | | | | | | | | |
| Immune response | 6 | 2% (1–5%) | 0 | – | 1 | 0% (0–2%) | 4 | 1% (0–4%) |
| Reciprocal antibody titers | 25.54 | (19.16–49.79) | – | – | 56.89 | (56.89–56.89) | 119.0 | (53.98–226.3) |
| Two doses, 14 weeks | | | | | | | | |
| Cumulative immune response | 6 | 2% (1–5%) | 25 | 9% (6–12%) | 1 | 0% (0–2%) | 11 | 4% (2–7%) |
| Reciprocal antibody titers | 11.31 | (9.58–11.31) | 22.63 | (14.22–28.44) | 22.63 | (22.63–22.63) | 28.44 | (20.31–67.88) |
| Three doses, 18 weeks | | | | | | | | |
| Cumulative immune response | 24 | 8% (5–12%) | 32 | 11% (8–15%) | 10 | 3% (2–6%) | 22 | 7 (5–11%) |
| Reciprocal antibody titers | 22.63 | (14.22–71.11) | 12.77 | (9.00–24.08) | 18.00 | (14.22–49.78) | 16.11 | (11.31–22.63) |
| **Type 3** | | | | | | | | |
| One dose, 10 weeks | | | | | | | | |
| Immune response | 10 | 3% (2–6%) | 1 | 0% (0–2%) | 4 | 1% (0–4%) | 236 | 79% (74–84%) |
| Reciprocal antibody titers | 1152 | (629.3-≥1448) | 910.2 | (910.2–910.2) | 938.1 | (571.5–1226) | 1152 | (576.0-≥1448) |
| Two doses, 14 weeks | | | | | | | | |
| Cumulative immune response | 17 | 6% (3–9%) | 15 | 5% (3–8%) | 8 | 3% (1–5%) | 273 | 92% (88–94%) |
| Reciprocal antibody titers | 724.1 | (288.0-≥1448) | 910.2 | (32.22-≥1448) | 650.1 | (343.5-≥1448) | 1152 | (576.0-≥1448) |
| Three doses, 18 weeks | | | | | | | | |
| Cumulative immune response | 28 | 9% (6–13%) | 22 | 8% (5–11%) | 18 | 6% (4–10%) | 285 | 96% (93–98%) |
| Reciprocal antibody titers | 743.1 | (181.0-≥1448) | 910.2 | (30.33-≥1448) | 376.1 | (76.63–1152) | 1024 | (455.1-≥1448) |

Data are the percentage of participants with immune response expressed as n/N and percentage including 95% confidence interval (CI). Immune response defined as seroconversion from seronegative (<1:8) to seropositive (≥1:8), or a four-fold rise in antibody titers among seropositives adjusted for maternal antibody decay, and seropositivity maintained through 18 weeks of age. Cumulative immune response was defined as immune response at any point up to and including at the time of assessment. Reciprocal antibody titers are presented as the median (interquartile range) among responders. mOPV1 = monovalent oral poliovirus vaccine type 1. fIPV = fractional inactivated poliovirus vaccine. bOPV = bivalent oral poliovirus vaccine.

## Table 5

Summary of reported adverse events among participants by study arms.

| | Arm A mOPV1 + fIPV6 (n = 301) | | Arm B mOPV1 + fIPV10 (n = 295) | | Arm C mOPV1 (n = 298) | | Arm D bOPV (n = 298) | |
|---|---|---|---|---|---|---|---|---|
| Any adverse events (AE) | 26 | 9% | 17 | 6% | 30 | 10% | 17 | 6% |
| Fever | 1 | 0% | 1 | 0% | 1 | 0% | 4 | 1% |
| Chickenpox | 1 | 0% | 0 | 0% | 1 | 0% | 0 | 0% |
| Gastrointestinal-related | 1 | 0% | 4 | 1% | 5 | 2% | 4 | 1% |
| Meningitis | 0 | 0% | 0 | 0% | 0 | 0% | 1 | 0% |
| Respiratory - acute | 13 | 4% | 9 | 3% | 20 | 7% | 8 | 3% |
| Respiratory - pneumonia | 9 | 3% | 6 | 2% | 8 | 3% | 2 | 1% |
| Other minor illness | 2 | 1% | 0 | 0% | 0 | 0% | 1 | 0% |
| Any serious adverse events (SAE) | 6 | 2% | 10 | 3% | 9 | 3% | 4 | 1% |

Data are n participants and % of participants in assigned arm. mOPV1 = monovalent oral poliovirus vaccine type 1. fIPV = fractional inactivated poliovirus vaccine. bOPV = bivalent oral poliovirus vaccine.