ORIGINAL ARTICLE

A Survey to Assess Serological Prevalence of Poliovirus Antibodies in Areas With High-Risk for Vaccine-Derived Poliovirus Transmission in Chad

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Background. World Health Organization African region is wild poliovirus-free; however, outbreaks of vaccine-derived poliovirus type 2 (VDPV2) continue to expand across the continent including in Chad. We conducted a serological survey of polio antibodies in polio high-risk areas of Chad to assess population immunity against poliovirus and estimate the risk of future outbreaks.

Methods. This was a community-based, cross-sectional survey carried out in September 2019. Children between 12 and 59 months were randomly selected using GIS enumeration of structures. Informed consent, demographic and anthropometric data, vaccination history, and blood spots were collected. Seropositivity against all 3 poliovirus serotypes was assessed using a microneutralization assay at Centers for Disease Control and Prevention, Atlanta, GA, USA.

Results. Analyzable data were obtained from 236 out of 285 (82.8%) enrolled children. Seroprevalence of polio antibodies for serotypes 1, 2, and 3 was 214/236 (90.7%); 145/236 (61.4%); and 196/236 (86.2%), respectively. For serotype 2, the seroprevalence significantly increased with age ($P = .004$); chronic malnutrition was a significant risk factor for being type 2-seronegative.

Interpretation. Poliovirus type 2 seroprevalence in young children was considered insufficient to protect against the spread of paralytic diseases caused by VDPV2. Indeed, VDPV2 outbreaks were reported from Chad in 2019 and 2020. High-quality immunization response to these outbreaks is needed to prevent further spread.

Key words: antibodies; Chad; polio; seroprevalence.

Currently, 5 out of the 6 World Health Organization (WHO) regions have been certified wild poliovirus-free [1, 2], including the African region, where the last case of wild poliovirus type 1 (WPV1) was detected in Nigeria in August 2016 [3]. However, complete polio eradication involves eradication of all polioviruses including those emanating from the use of live oral poliovirus vaccine (OPV) in recipients or their close contacts, referred to as vaccine-associated paralytic poliomyelitis or by genetic reversion through extensive replication, referred to as vaccine-derived polioviruses (VDPVs) [4]. In rare circumstances, the extensive transmission of Sabin-like strains in under-immunized communities allows genetic reversion that leads to outbreaks of

paralytic poliomyelitis due to circulating VDPVs (cVDPVs) [5]. Therefore, the continuous use of Sabin strain OPV is not compatible with poliovirus eradication; OPV needs to be ultimately withdrawn and gradually replaced by inactivated poliovirus vaccine (IPV) [6]. The first phase of withdrawal was the global removal of the Sabin type 2 strain from trivalent OPV (tOPV), switching to bivalent OPV (bOPV; types 1 and 3) in April 2016 [7] for all immunization activities. Outbreaks of cVDPVs have been increasing in recent years [8], predominantly of serotype 2 (cVDPV2) [9], of which, 77% of cases from July 2018 to March 2020 were on the African continent [8]. While many outbreaks after the switch were controlled promptly, some have taken multiple supplementary immunization activities using monovalent OPV type 2 (mOPV2) to control or stop spread and some continue. New emergent cVDPV2 outbreaks, seeded during low-quality mOPV2 outbreak responses, are also on the rise as the time since the switch has progressed.

Sub-Saharan African countries have experienced 49 outbreaks of cVDPV2 between April 2016 (when tOPV was replaced by bOPV) and the end of 2020 [8]. There were 3 outbreaks reported from Chad—one cVDPV2 importation from Nigeria in 2019, a new emergence of cVDPV2 detected in Chad in 2019, and one importation from Central African

Republic in 2020 [10]. Chad responded to the cVDPV2 outbreak with mOPV2 campaigns. One such campaign targeting all children below 5 years of age was conducted in July 2019, just prior to this survey. Most of the cases in Chad were detected in the western and southern areas of the country considered as high risk.

Chad introduced 1 dose of IPV in routine immunization, in 2015 and continues to administer bOPV in their immunization schedule (the routine immunization schedule includes bOPV at birth, 6, 10, and 14 weeks of age; and IPV at 14 weeks of age). The estimated national vaccination coverage in Chad with IPV and the third bOPV dose has been around 50% [11].

Seroprevalence surveys have been serving as a tool to assess population immunity gaps. Several seroprevalence surveys carried out in Nigeria, other West African countries, India, and Pakistan have well demonstrated their utility [12–14]. To better understand the immunity in areas and populations at high risk of polio in Chad and to assess the risk of future paralytic outbreaks of VDPVs, we conducted a community-based poliovirus seroprevalence survey in Chad. In our analysis, we focused primarily on type 2 seroprevalence because of the risk of cVDPV2 outbreaks.

METHODS

A community-based, cross-sectional survey was conducted in September 2019 that included areas of Hadjer-Lamis, Lac, and Kanem provinces of Chad. These selected areas have a great inflow of internally displaced persons and refugees from neighboring Nigeria, Niger, and the conflict areas of the Lake Chad region due to the Boko Haram and other rebel groups' insurgency; thereby, they were considered as high-risk areas of Chad.

Using ArcGIS software, a sampling frame of settlements in these selected provinces was obtained. Settlements were plotted onto a map over satellite imagery and structures or households were manually digitized within these settlements. Households were then randomly selected within each of the settlements using computer-generated random numbers. One child, 12-59 months of age, living in each candidate household was eligible to be enrolled in the survey. If a household had 2 or more children aged 12-59 months, only one child was randomly selected using dice or other means.

A sample size of 334 children was calculated in order to detect, at a 95% confidence level, a seroprevalence point estimate with a precision of approximately ±5% assuming >80% seroprevalence and the proportion of non-consenting parents <10%. Assuming that in approximately 2 out of 3 identified structures there would be at least 1 child in the targeted age group, 500 households were planned to be selected for inclusion in the study. GPS coordinates of the visited households were recorded. We assumed that seroprevalence will be distributed homogeneously across this area.

A simple demographic questionnaire including poliovirus vaccination history was administered to the parent or legal guardian, and children's weight and height were recorded. Acute and chronic malnutrition was defined as weight-for-age Z-score ≤-2 and height-for-age Z-score ≤-2 standard deviations from mean Z-score, respectively. Vaccination card was requested, and documented vaccination history was recorded; if a vaccination card was available or not, the caretaker was asked if mOPV2 was received during a campaign in 2019. Trained phlebotomists collected dried blood spot (DBS) on Whatman 903tm cards using a finger prick technique. The DBS cards were shipped to the Centers for Disease Control and Prevention (CDC) in Atlanta, GA, USA and tested for the presence of poliovirus neutralizing antibodies using a standard neutralization assay. Seroprevalence or seropositivity was defined as reciprocal titers of poliovirus neutralizing antibodies >8, with the highest reported antibody titer of >1:1448 (no further dilution made). Titers were reported in $\log_2$ scale [15].

Seroprevalence against all 3 poliovirus types was expressed as percentages with 95% confidence intervals. Risk factors for type 2 seroprevalence were assessed using Fisher's exact test or chi-square test, as appropriate. Cochrane-Armitage trend test was applied to test for trend in seroprevalence by age of the child. A multivariable logistic regression was applied using factors that were significant at 0.25 level in the bivariate analysis or unadjusted analysis for type 2 seroprevalence. The reciprocal titers were presented using median for all 3 serotypes. A P value <.05 was considered as significant. The data were analyzed using R software (v.3.4.3).

The study protocol was reviewed and approved by the Chad's National Committee of Bioethics and by WHO's Ethics Review Committee.

RESULTS

A total of 285 children were enrolled from 727 visited households. In 2 of the 285 children, blood was not collected due to refusal; in 47 of 285 children, the collected blood was not sufficient in volume for analysis, leading to the study population of 236, representing 83% of enrolled children.

Sociodemographic indicators and vaccination histories are presented in Tables 1 and 2, respectively. Nomadic population constituted about 4% (10/236) of the study population. Most parents indicated that their child received mOPV2 dose in a vaccination campaign in July 2019 (215/236, 91.5%, Table 2). Only 8% (19/236) of families presented an immunization card. Of those with the vaccination card, 79% (15/19) had received third dose of OPV and 61.1% (11/18) received IPV. Among the enrolled children in the survey, 37% (137/218) had chronic malnutrition while 20% (44/218) were acutely malnourished.

Seroprevalence was 91% (95%CI: 86.2-94.1) (214/236), 61% (95%CI: 54.9-67.7) (145/236), and 83% (95%CI: 77.6-87.6)

Table 1. Sociodemographic Indicators of the Study Population

Indicators		n/N	%
Gender	Female	120/236	50.9
Age	12-23 months	30/236	12.7
	24-35 months	63/236	26.7
	36-47 months	83/236	35.2
	48-59 months	60/236	25.4
Population type	Resident	216/236	91.5
	Nomads	11/236	4.7
	Internally displaced persons	9/236	3.8
Location type	Village	211/236	89.4
	Internally displaced people camp	9/236	3.8
	Nomadic settlement	10/236	4.2
	Town	6/236	2.5
Type of toilet	Open air	191/236	80.9
	Communal latrines	42/236	17.8
	Private latrines/toilettes	3/236	1.3
Water source	Village pump	193/236	81.8
	River	24/236	10.2
	Stagnant water	3/236	1.3
	Running water	3/236	1.3
	Other	13/236	5.5

Table 2. Vaccination History and Nutritional Status

Indicator	n/N	%
Vaccination history		
Vaccination card available	19/236	8.1
mOPV2 received in campaign in 2019 prior to the survey	215/236	91.5
Third dose of OPV received in routine immunization (card only)	15/19	79.0
IPV received (card only)	11/18	61.1
First dose of measles vaccine received (card only)	7/17	41.2
Second dose of measles vaccine received (card only)	0/13	–
Nutritional status		
Chronic malnutrition (≤-2 Z-score height-for-age)	137/218	37.2
Acute malnutrition (≤-2 Z-score weight-for-age)	44/218	20.1

Abbreviations: IPV, inactivated poliovirus vaccine; mOPV2, monovalent OPV type 2; OPV, oral poliovirus vaccine.

(196/236) against poliovirus types 1, 2, and 3, respectively (Figure 1). The seroprevalence was stratified by age of the child categorized into 4 groups of 12 months each from 12 to 59 months. There was a significant increasing trend in seroprevalence with increase in the age of a child for serotypes 1 (P = .003) and 2 (P < .001). For type 1 seroprevalence was 86.7% (95%CI: 68.2-100) for children 12-23 months compared to 98.3 (95%CI: 91.1-99.9) for 48-59 months; for type 2, it was 36.7% (95%CI: 19.2-54.3) for 12-23 months compared to 83.3% (95%CI: 71.8-92.4) for 48-59 months.

The median reciprocal titers of antibodies were significantly different across the age groups for serotype 2 only (P = .006) (Figure 2). The cumulative distribution plots for titers of all 3 types in $\log_2$ scale are presented in Figure 3.

The design effect was calculated to be 1.01 indicating homogenous distribution of seroprevalence.

Bivariate analysis of risk factors for type 2 seronegativity is presented in Table 3. Risk factors included in multivariable regression (Table 4) were gender, age, chronic malnutrition, being born before the switch from tOPV to bOPV vaccine, and type of toilet. The odds of type 2 seropositivity were 3 times higher among those children born before switch as compared to children born after switch (OR = 3; 95% CI: 1.56-7.14). Children with chronic malnutrition were less likely to be type 2 seropositive (OR = 0.55; 95% CI: 0.31-0.98).

DISCUSSION AND CONCLUSION

Routine immunization national coverage estimates (which don't include doses given during supplementary campaigns) for the third dose of polio in Chad have been sub-optimal since 1990 until the latest available estimate for 2019 [11]. Yet the seroprevalence of antibodies against poliovirus of types 1 and 3 was found to be high in our survey—91% and 83%,
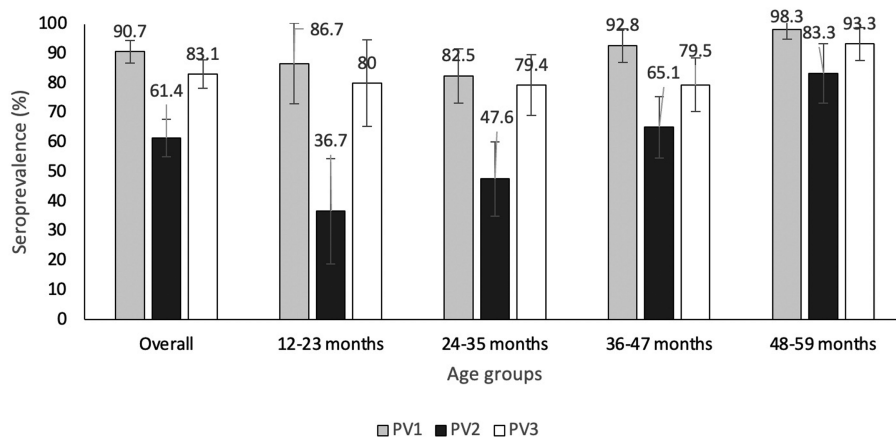
Figure 1. Overall and age groups wise seroprevalences against all 3 poliovirus types.
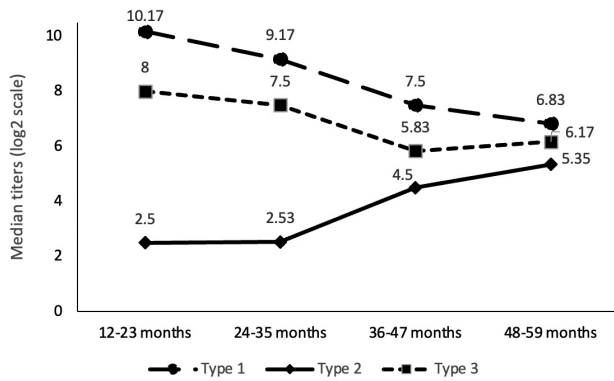
Figure 2. Median poliovirus type-specific titers (in $\log_2$ scale) by age.

Figure 3. Reverse cumulative distribution plots of reciprocal titers in $\log_2$ scale against all 3 poliovirus types.

respectively. This is likely a result of 14-16 rounds of bOPV supplementary immunization campaigns carried out in Chad since the April 2016 replacement of tOPV with bOPV vaccine, no IPV campaigns have been conducted during this period. In contrast, type 2 seroprevalence was low overall (61%). Children born after the switch had limited opportunity to receive type 2 containing vaccine—one opportunity was during the mOPV2 campaign conducted in July 2019, the other opportunity was through IPV provided in routine

Table 3. Association of Sociodemographic, Vaccination History, and Nutritional Status With Type 2 Seroprevalence—Univariate Analysis

	Seropositive		
Indicators	n	%	P value
Gender			
Female	69	57.5	.230
Male	76	65.5	
Born before or after bOPV to tOPV switch (April 2016)			
After	95	54.0	<.001*
Before	50	83.3	
Age			
<2 years	11	36.7	.004*
≥2 years	134	65.0	
Population type			
Nomads	5	45.5	.344
Others	140	62.2	
Vaccination history			
Received IPV	8	72.7	.627
Not received IPV	4	57.1	
Chronic malnutrition			
Stunted	48	52.7	.039*
Normal	97	66.7	
Acute malnutrition			
Wasted	29	58.0	.625
Normal	113	62.1	
Toilet			
Open field	125	65.4	.011*
Latrine	20	44.4	

Abbreviations: bOPV, bivalent oral poliovirus vaccine; IPV, inactivated poliovirus vaccine; tOPV, trivalent oral poliovirus vaccine.
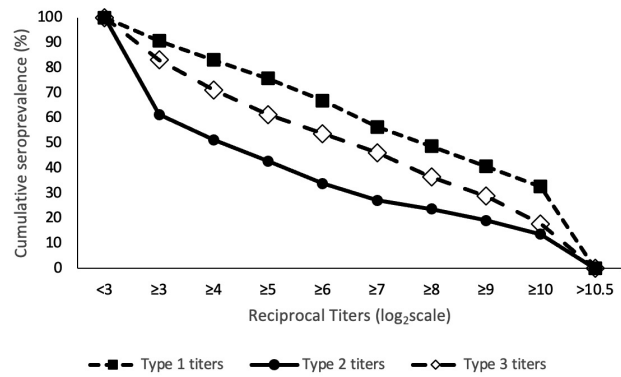
*Significant at $P < .05$.

immunization, as well as possible exposure for some children to cVDPV2 transmission. The identified type 2 immunity gap likely was present in non-sampled areas and greatly contributed to the spread of cVDPV2 in late 2019 and 2020 across the region.

The risk of future cVDPV2 outbreaks and spread remains high; and as time since the switch to bOPV increases, population immunity to type 2 polio continues to decrease [8, 16]. Chad shares borders with countries that recently reported type 2 outbreaks (Nigeria, Cameroon, Niger, and Central African Republic), and population movement between these countries and Chad is well documented [17, 18]. Exacerbating the situation is the ongoing insecurity due to Boko Haram insurgency resulting in the disruption of vaccination services and population displacement [17–19]. This increases the chance of cVDP2 importation again into Chad or from Chad into neighboring countries and further international transmission. Lastly, Chad has areas of insecurity which limits the quality of response SIAs in those areas.

This survey demonstrated association between seronegativity and chronic malnutrition. This finding is in agreement with previously reported results of studies in Pakistan [14], Morocco [20], and India [13].

Our study had some limitations. A long delay in laboratory testing of the DBS samples made it difficult to provide timely feedback to the polio program in Chad. Furthermore,

Table 4. Risk Factor Analysis of Type 2 Seroprevalence—Multivariate Analysis

Risk Factors	aOR	95% CI	P value
Gender (female vs male)	0.80	0.46-1.41	.438
Switch (April 2016) (born after vs before)	0.30	0.14-0.64	.002*
Age (<2 years vs ≥2 years)	0.45	0.19-1.05	.064
Chronic malnutrition (stunted vs normal)	0.55	0.31-0.98	.042*
Toilet (open field vs latrine)	1.79	0.88-3.63	.108

aOR—adjusted odds ratio for gender, age, born before/after the switch, malnutrition, and type of toilet.

*Significant at $P < .05$.

we have not collected vaccination history by recall and therefore we have vaccination history only from a very small subset of children whose parents kept the vaccination cards (~8%). In reporting the total number of OPV doses received, we were unable to distinguish what type of OPV was administered. We estimated sample size assuming type 2 seroprevalence of 80%, however, in our sample, it was 61% which may have resulted in wider confidence intervals than what we had originally intended.

Our survey provides further evidence that poor routine immunization coverage may lead to spread of paralytic disease caused by VDPV if importation events or seeding of VDPVs occurs.

Notes

References

1. CDC Global Health – Polio – Our Progress [Internet]. **2019**. Accessed February 5, 2021. https://www.cdc.gov/polio/progress/index.htm
2. Global Polio Eradication Initiative Applauds WHO African Region for Wild Polio-Free Certification [Internet]. Accessed February 5, 2021. https://www.who.int/news/item/25-08-2020-global-polio-eradication-initiative-applauds-who-african-region-for-wild-polio-free-certification
3. Leke RGF, King A, Pallansch MA, et al. Certifying the interruption of wild poliovirus transmission in the WHO African region on the turbulent journey to a polio-free world. Lancet Glob Health **2020**; 8(10):e1345–51.
4. GPEI-Our Mission [Internet]. Accessed February 5, 2021. https://polioeradication.org/who-we-are/our-mission/
5. Circulating Vaccine-Derived Poliovirus – GPEI [Internet]. Accessed February 24, 2021. https://polioeradication.org/polio-today/polio-now/this-week/circulating-vaccine-derived-poliovirus/
6. GPEI: Polio Endgame Strategy 2019–2023. Eradication, Integration, Certification and Containment [Internet]. Accessed February 23, 2021. https://apps.who.int/iris/bitstream/handle/10665/329948/WHO-Polio-19.04-eng.pdf?sequence=1
7. GPEI-Global Synchronisation and the Switch [Internet]. Accessed February 5, 2021. https://polioeradication.org/news-post/global-synchronisation-and-the-switch/
8. Alleman MM, Jorba J, Greene SA, et al. Update on vaccine-derived poliovirus outbreaks – worldwide, July 2019-February 2020. MMWR Morb Mortal Wkly Rep **2020**; 69:489–95.
9. Jorba J, Diop OM, Iber J, et al. Update on vaccine-derived poliovirus outbreaks – worldwide, January 2018-June 2019. MMWR Morb Mortal Wkly Rep **2019**; 68:1024–8.
10. WHO. Circulating Vaccine-Derived Poliovirus Type 2 – African Region. Emergencies Preparedness, Response [Internet]. **2019**. https://www.who.int/csr/don/29-november-2019-polio-african-region/en/. Accessed May 24, 2021.
11. WHO and UNICEF. Chad: WHO and UNICEF Estimates of Immunization Coverage: 2019 Revision. **2020**. https://www.who.int/immunization/monitoring_surveillance/data/tcd.pdf?ua=1. Accessed May 24, 2021.
12. Gamage D, Palihawadana P, Mach O, Weldon W, Oberste S, Sutter R. Achieving high seroprevalence against polioviruses in Sri Lanka—results from a serological survey, 2014. J Epidemiol Glob Health **2015**; 5:S67–71.
13. Bahl S, Estívariz CF, Sutter RW, et al. Cross-sectional serologic assessment of immunity to poliovirus infection in high-risk areas of northern India. J Infect Dis **2014**; 210(Suppl 1):S243–51.
14. Yousuf A, Syed Shah SA, Syed Jaffery IA, et al. Seroprevalence rate of Poliovirus antibodies among the Healthy and Protein Energy Malnutrition children. Pak J Med Sci **2015**; 31:403–7.
15. Weldon WC, Oberste MS, Pallansch MA. Standardized methods for detection of poliovirus antibodies. Methods Mol Biol **2016**; 1387:145–76.
16. Macklin GR, O'Reilly KM, Grassly NC, et al. Evolving epidemiology of poliovirus serotype 2 following withdrawal of the serotype 2 oral poliovirus vaccine. Science **2020**; 368:401–5.
17. Oginni O, Opoku M, Alupo B. Terrorism in the Lake Chad Region: integration of refugees and internally displaced persons. J Borderl Stud **2018**; 35:725–41.
18. Bawa S, Afolabi M, Abdelrahim K, et al. Transboundary nomadic population movement: a potential for import-export of poliovirus. BMC Public Health **2018**; 18:1316.
19. Awosusi AE. Aftermath of Boko Haram violence in the Lake Chad Basin: a neglected global health threat. BMJ Glob Health **2017**; 2:e000193.
20. Caidi H, Bennis IF, Mouan N, El Aouad R. [Evaluation of the response to vaccination against poliomyelitis and measles in malnourished children in Morocco]. East Mediterr Health J **2004**; 10:474–81.