Title

Population and Geospatial Risks of Vaccine-Derived Poliovirus Type-2 in the Democratic Republic of the Congo

Author

Halbrook, Megan

Publication Date

2021

Peer reviewed|Thesis/dissertation

UNIVERSITY OF CALIFORNIA

Los Angeles

Population and Geospatial Risks of Vaccine-Derived Poliovirus Type-2 in the Democratic Republic of the Congo

A dissertation submitted in partial satisfaction of the requirements for the degree Doctor of Philosophy in Epidemiology

by

Megan Halbrook

2021

ABSTRACT OF THE DISSERTATION

Population and Geospatial Risks of Vaccine-Derived Poliovirus Type-2 in the Democratic Republic of the Congo

by

Megan Halbrook

Doctor of Philosophy in Epidemiology

University of California, Los Angeles, 2021

Professor Anne W. Rimoin, Chair

Poliovirus, once a global pandemic is now in its endgame eradication stages. Currently, wild poliovirus type 2 and 3 have been declared eradicated by the World Health Organization (WHO) and wild type 1 is circulating in just two remaining countries. As the Sabin oral polio vaccine utilizes a live attenuated strain of poliovirus, in 2012 the WHO General Assembly released a strategic plan for polio eradication that called for the eventual removal of the oral polio vaccine. In April 2016, the WHO coordinated a global switch day where trivalent oral polio vaccines containing poliovirus type 2 were replaced with bivalent vaccines containing polio types 1 and 3 only. Since then, vaccine-derived poliovirus infections in the Democratic Republic of the Congo (DRC) have risen as polio vaccine coverage rates, and specifically rates against serotype 2, have fallen. This dissertation serves to contribute towards polio eradication efforts in the DRC by describing the landscape of childhood vaccination against poliovirus in a vaccine-derived

poliovirus endemic region and quantify how the key vaccine intervention, supplementary immunization campaigns, contribute to improving vaccine coverage. Chapter 1 provides a summary of the pathology of polio and its pandemic history, as well as the WHO endgame strategic plan and policies and the landscape of polio in DRC. Chapter 2 quantifies community immunity against poliovirus and risk factors for under immunization among children under five in an outbreak prone region of southeastern DRC. Chapter 3 explores how repeated immunization interventions impacts community vaccine coverage and uses propensity score weighting to compare a gold standard biomarker for vaccination to material recall collected via questionnaire. Chapter 4 explores the spatial point process of vaccine-derived poliovirus cases and how spatial access to health care can impact under-vaccination. This dissertation finds that ultimately, access to vaccine in the DRC is an enduring issue related to challenges associated with DRC's resource-poor health system and systemic issues with infrastructure. Vaccine immunization campaigns do improve community immunity, but likely are not providing adequate coverage to halt the spread of vaccine-derived polio viruses. Addressing the limitations of the current vaccine strategy head on can help move the DRC closer to polio eradication.

The dissertation of Megan Halbrook is approved.

Sudipto Banerjee

Robert J. Kim-Farley

Roch A. Nianogo

Anne W. Rimoin, Committee Chair

University of California, Los Angeles

2021

To my family and friends, thank you for years of support and encouragement.

To the families of Haut Lomami and Tanganyika province that graciously agreed to participate in these studies, thank you.

TABLE OF CONTENTS

List of Tables

List of Figures

List of Acronyms

AFP	acute flaccid paralysis
DRC	Democratic Republic of the Congo
EPI	Expanded Programme on Immunization
GPEI	Global Polio Eradication Initiative
GPLN	Global Polio Laboratory Network
IPV	inactivated polio vaccine
NID	National immunization day
OPV	oral polio vaccine
bOPV	bivalent oral polio vaccine
mOPV	monovalent oral polio vaccine
tOPV	trivalent oral polio vaccine
PAHO	Pan American Health Organization
SIA	supplementary immunization act ivy
SNID	Subnational immunization day
UNICEF	United Nations Children's Fund
VAPP	vaccine-associated paralytic polio
VDPV	vaccine-derived polio virus
aVDPV	ambiguous vaccine-derived polio virus
cVDPV	circulating vaccine-derived polio virus
cVDPV2	circulating vaccine-derived polio virus type 2
iVDPV	immunodeficiency-related vaccine-derived polio virus
WHO	World Health Organization
WPV	wild polio virus
WPV1	wild polio virus type 1
WPV2	wild polio virus type 2
WPV3	wild polio virus type 3

ACKNOWLEDGMENTS

A heartfelt thank you goes out to my doctoral advisor, Anne Rimoin, who has supported my academic progress and my desire for adventure and excitement throughout this program. I've been able to achieve goals I've long dreamt about and I leave UCLA with many fun stories thanks to this program you've created.

My time in Congo will forever be some of my most cherished memories and to the folks there—Nicole Hoff, Kamy Musene, Camille Dzogang, Francine Miole, Merly Tambu, Fabrice Bito, Michael Beia, and Millet—thank you so much for welcoming me into your world and for your patience while 'spoke' French.

I'd like to thank my other colleagues and classmates who have offered support, guidance, and solidarity throughout this process especially Dr. Kirsty Clark and Dr. Adva Gadoth. Kirsty—your friendship was an eternal summer during what felt like an eternal winter, it's rare to find someone both absolutely hilarious and absolutely brilliant. Adva—you are not only my research partner but dearest friend and confidant, we've been lockstep since day 1, and I owe much of my success to your friendship. I'd also like to thank Dr. Rick Shoenburg for his support, both academic and moral, and for volunteering his time and guidance on the point process methods used in Chapter 4.

Finally, I am deeply grateful for the support from my family members and the patience you have shown me while I "go at my own pace". I'm happy public health science is something we can all talk about together. To Jonatán López, thank you so much for your patience and understanding and listening. Thank you for the postmates and the constant encouragement.

VITA

Education:

Columbia University Mailman School Master of Public Health, Infectious Disease Epidemiology	New York, NY May 2016
McGill University Bachelor of Arts- Medical Anthropology	Montreal, Canada April 2011

Teaching Experience:

University of California, Los Angeles Teaching Fellow Teaching Associate	Los Angeles, CA September 2019- June 2020 September 2016 – June 2019
Columbia University Mailman School Teaching Assistant	New York, NY September 2015- May 2016

Selected Publications:

Gadoth A, **Halbrook M**, Martin-Blais R, Gray A, Tobin NH, Ferbas KG, Aldrovandi GM, Rimoin AW. Assessment of COVID-19 vaccine acceptance among healthcare workers in Los Angeles

medRxiv 2020.11.18.20234468; doi: https://doi.org/10.1101/2020.11.18.20234468. *Pre-print, under review.*

Halbrook M, Gadoth A, Martin-Blais R, Gray A, Contreras D, Kashani S, Kazan C, Kane B, Tobin NH, Fulcher JA, Brooker SL, Kitchen SG, Faure-Kumar E, Yang OO, Ferbas KG, Aldrovandi GM, Rimoin AW. Incidence of SARS-CoV-2 infection among asymptomatic frontline health workers in Los Angeles County, California. MedRxiv 2020.11.18.20234211; doi: https://doi.org/10.1101/2020.11.18.20234211. *Pre-print*

Halbrook M, Gadoth A, Shankar A, Zheng H, Campbell E, Hoff NA, Muyembe JJ, Peeters M, Okitolonda Wemakoy E, Rimoin AW, Switzer WM. Human T-cell Lymphotropic Virus Type 1 Transmission Dynamics in Rural Villages in the Democratic Republic of the Congo with High Nonhuman Primate Exposure. PLOS NTD. *In Press.*

Alfonso VH, **Halbrook M**, Hoff NA, Voorman A, Mukadi P, Weldon WC, Gadoth A, Sinai C, Musene K, Spencer D, Gerber S, Mukadi D, M Bwaka A, Ngoie Mwamba G, Muyombi E, Muyembe JJ, Okitolonda-Wemakoy E, Rimoin AW. Vaccination and serologic response against poliovirus among children in an outbreak area of vaccine derived poliovirus in the Democratic Republic of the Congo. *Under review.*

Bratcher A, Hoff N, Doshi R, Gadoth A, **Halbrook M**, Mukadi P, Musene K, Kebela Ilunga B, Spencer D,

Bramble M, McIlwain D, Kelly, J, Mukadi, D, Ahuka-Mundeke S, Okitolonda E, Muyembe JJ, Rimoin, AW. Seroprevalence of Ebola virus GP antibodies by zoonotic exposures in the Democratic Republic of Congo. *Under review.*

Vasquez AM, Lake J, Ngai S, **Halbrook M**, et al. Notes from the Field: Fungal Bloodstream Infections Associated with a Compounded Intravenous Medication at an Outpatient Oncology Clinic — New York City, 2016. MMWR Morb Mortal Wkly Rep 2016;65:1274–1275. DOI: http://dx.doi.org/10.15585/mmwr.mm6545a6.

Weiss, D., Boyd, C., Rakeman, J. L., Greene, S. K., Fitzhenry, R., McProud, T., ... & Fine, A. D. (2017). A large community outbreak of Legionnaires' disease associated with a cooling tower in New York City, 2015. Public health reports, 132(2), 241-250.

Danielle Bloch, Kenya Murray, Eric Peterson, Stephanie Ngai, Inessa Rubinstein, Tanya A Halse, Ifeoma Ezeoke, Laura Miller, Lola Arakaki, Arianne Ramautar, Mike Antwi, Paula Del Rosso, Marie Dorsinville, Sandhya Clark, **Megan Halbrook**, Joseph Kennedy, Sarah Braunstein, Don Weiss; Sex Difference in Meningococcal Disease Mortality, New York City, 2008–2016, Clinical Infectious Diseases, Volume 67, Issue 5, 16 August 2018, Pages 760–769, https://doi.org/10.1093/cid/ciy183

Parton, H.B., Mathes, R., Abdelnabi, J., Alleyne, L., Econome, A., Fitzhenry, R., Forney, K., **Halbrook, M.**, Ngai, S. and Weiss, D., 2016. Investigating a Syndromic Surveillance Signal with Complimentary Data Systems. Online Journal of Public Health Informatics, 8(1). DOI: https://doi.org/10.5210/ojphi.v8i1.6569

Chapter 1. Introduction and Background

1.1 Polio Epidemiology

First discovered in 1908, polioviruses are neurotropic RNA group C enteroviruses of the *Picornaviridae* family naturally found solely in human reservoirs and transmitted between persons through feces or pharyngeal secretions via the fecal-oral or oral-oral route.[1-4] The resulting disease, poliomyelitis, is caused by one of three poliovirus antigenic types (types 1, 2, or 3) based on their reaction with reference panels of neutralizing antisera.[5,6] These three serotypes differ in virulence and scope of individuals infected—the paralytic attack rate is greatest with type 1 (~0.5%) and is the least for type 2 (<0.05%)[7]—though transmission dynamics of each wild-type are roughly similar. A 1999 meta-analysis[8] observed that the R_0 for wild poliovirus (WPV) ranged from 2-4 in industrialized areas with good hygiene, and from 8-14 in areas with poor hygiene. Infectious virus can be found in 92-97% of persons with a primary infection. The virus can shed via the oropharynx from around one to three weeks and via feces for an average of five to eight weeks.[9] However, the duration of infectiveness has been observed to be as long as four months. In an infection among immunocompromised individuals viremia can last up to several years.[8]

Due to the transmission of polio via the fecal-oral route, environmental persistence of polioviruses is of interest. In all societies, fecal contamination is ubiquitous. In regions with poor water, sanitation, and hygiene (WASH) or households with children under five years of age the risk of fecal contamination rises. WPVs are more robust than OPV strains but both polioviruses are sensitive: high temperatures and ultraviolet exposure will cause inactivation.[10]

Polio is best known for causing paralysis but in fact, nearly 90-95% of cases are asymptomatic. Among children, 72% of cases are asymptomatic and 24% of cases consist of

abortive poliomyelitis, a mild disease lasting around a week characterized by various nonspecific symptoms including vomiting, headache, fever, constipation, fatigue, and sore throat. Among more serious cases, poliovirus can present as aseptic meningitis (4%).[11] For non-paralytic poliomyelitis the incubation period is around 4-6 days. Paralytic poliomyelitis presents as acute flaccid paralysis (AFP) and occurs in <1% (0.5-1%) of infected individuals.[1,12,13] Prior to acute onset of paralysis, a minor illness followed by several asymptomatic days is commonly observed.[7] The incubation period for symptom onset in paralytic cases is usually seven to 21 days and following early signs of infection paralysis can begin one to 18 days later.[11] On average, the incubation period from infection to paralysis is around 10 days.[12,14,15]

Paralytic poliomyelitis is classified into three types. The most common is spinal polio which is characterized by asymmetrical paralysis of the lower limbs. Bulbar polio is more often observed in children and is associated with paralysis of the nerves that control the muscles in the neck and tongue. Bulbospinal polio affects the C3-5 spinal cord which inhibits the function of the diaphragm and results in death.[16,17] Cumulatively, the WHO estimates that 10-20 million persons have been impacted by paralytic poliomyelitis.[18] AFP is characterized by fever and rapid onset of muscle weakness that can progress to maximum severity in a little as a few day to weeks. AFP represents a significant morbidity for poliovirus as paralysis can last a lifetime, but also a high mortality risk as respiratory and esophageal muscles can become paralyzed. Case fatality rates (CFR) among AFP cases is around 5-10%, increasing with age of illness.[13] CFR among children is 2-5%, and among adults is between 15-30% depending on age of onset. Poliomyelitis cases with bulbar paralysis can have a CFR between 25-75%.[11] Following an infection, lifelong serotype specific immunity is conferred. Many cases of paralytic polio recover muscle function at least partially however any weakness or paralysis lasting 6-12 months after

onset is usually permanent.[11,19] Wild poliovirus type 1 (WPV1) is the most neurovirulent and has the highest proportion of paralytic infection.[13]

Major risk factors for poliovirus transmission include poor sanitation and hygiene, tropical and sub-tropical conditions, large birth cohorts, and high population densities.[7,20,21] Available studies provide weak evidence that the rates of paralytic cases increase with age of infected individual, but do demonstrate that among paralytic cases there is a marked age-specific increase from infancy in the probability of severe paralysis.[12,20] Understanding the distribution of poliovirus infections across age groups is important to understanding the burden of poliomyelitis. A recent study in Pakistan suggests that poor immunity due to early childhood malnutrition may be a contributing factor towards persistent poliovirus circulation.[22] As most countries have eradicated wild poliovirus and there exists two highly effective poliovirus vaccines, today the risk factors for poliomyelitis are primarily socio-demographic factors that are associated with low vaccination rates.

1.2 Virology & Pathogenesis

Polioviruses small are a nonenveloped, single stranded RNA virus with approximately 7500 nucleotides.[3] Much of the genetic code is shared with other enteroviruses, some of which can also cause AFP but the four capsid proteins (VP1-4) are unique to poliovirus. Peptide loops extending from VP1, VP2, and VP3 form neutralizing antigenic sites which allow for easy laboratory identification and serological detection.[23,24] These neutralizing antigenic sites vary across poliovirus serotype requiring a type-specific antisera, but the range of variation is small such that polio vaccines can provide protective immunity to all antigenic variations.[7] Polioviruses target human cells via a specific poliovirus receptor, CD155.[25,26] After entry,

infected cells can show cytopathic effects within 4-6 hours. Upon cell death up to 10,000 infectious virus particles can be released.

Following ingestion of WPV, poliovirus can replicate in the pharynx, tonsils, Peyer's patches, and gastrointestinal mucosa.[3,17] During this period, transmission occurs primarily though viral shedding via feces. A meta-analysis on the duration of WPV excretion found that in a high proportion of persons poliovirus lasts 3-4 weeks but can persist up to 5-6 weeks in some cases. Duration of viral shedding was reduced in vaccinated individuals.[27] From the initial sites of replication, virus reaches mesenteric and cervical lymph nodes and then the blood, causing viremia. The majority of poliovirus infections are subclinical, and in these cases, infected individuals clear the virus experiencing mild symptoms. In some cases, viremia is maintained past this preliminary stage in extraneural tissue. In <1% of cases the virus will invade the central nervous system, attacking lower motor neurons causing acute flaccid paralysis. Poliovirus can invade the central nervous system either via the blood stream or through a peripheral nerve which then via axonal transport carries the virus to the central nervous system (Figure 1.1).[3,28]

1.3 Treatment and Prevention: Vaccines

The primary prevention method for poliovirus is immunization with the poliovirus vaccines. However, as poliovirus is transmitted primarily via the fecal-oral route behaviours that support proper water, sanitation, and hygiene practices (WASH) can also aid poliovirus prevention. Currently this is no cure for poliomyelitis, only supportive treatment to alleviate symptoms.

The inactivated polio vaccine (IPV), developed by Jonas Salk in 1955, contains all three poliovirus serotypes and is administered via injection. IPV initiates an IgG-mediated humoral immunity in the bloodstream which protects against paralysis but confers a weaker intestinal

immunity.[29,30] Thus, IPV does not interrupt shedding or halt the transmission cycle. However, challenge studies have shown that immunization with IPV can lead to a reduction in the duration or volume of viral shedding.[31-33]

The oral polio vaccine (OPV) was developed by Albert Sabin and used commercially beginning in 1961. Due to its oral administration, OPV protects an individual from paralysis and interrupts the oral-oral or fecal-oral transmission cycle. OPV can generate humoral immunity, oral mucosal immunity, and intestinal mucosal immunity and was crucial to the elimination of the global polio epidemic since the vaccine strain is easy to administer and can shed from a vaccinated person to an unvaccinated close contact.[8,34] This allowed for population immunity to increase at rates outpacing the vaccination rates.[8] Because of these immunological characteristics, the ease of administering an oral vaccine, and its low cost, OPV has facilitated polio eradication across much of the globe.[7] While OPV is easy and safe to administer, seroconversion rates are variable. Early studies identified that the three antigenic types of poliovirus in the OPV vaccine have their own seroconvergence rates and can interfere with each other, reducing immune response. Thus, in each type of OPV, tri-, bi-, or monovalent, the poliovirus serotype components will each have their own seroconvergence rates. In addition to the formation of the vaccine, lower seroconversion responses have been observed in the tropics compared to temperate climates. This can be attributed to several factors, including high level of maternal antibodies, interferences from concurrent enterovirus infections, and poor nutritional status.[8] Studies have found that the seroconversion rate was greater with monovalent OPV (mOPV) (65-95%) compared with trivalent OPV (tOPV) (50-90%). Studies have also shown that OPV2 produces high seroconversion rates, despite being less immunogenetic and in tOPV doses can delay the multiplication of types 1 and 3.[8] For these reasons, from 1961 to 1964 mOPV was

primarily used in the US until it was determined that the advantages of a single vaccine outweighed the small gain in seroconversion rates.[12] Additionally, a study by Manteen et al. suggests that bivalent OPV (bOPV) leads to significantly more seroconversion than tOPV.[35,36] The true seroconvergence rate for OPV depends heavily on vaccine type, immunological makeup of the population, and the timing and number of vaccine doses. The study by Manteen et al. conducted a literature review of OPV and IPV blood from umbilical cord draws among newborns who have received just the birth dose. A more contemporary study by Bandyopadhyay et. al. explored eight RCTs which used full vaccine regimens as their treatment groups. This metastudy found that despite varying regimens of bOPV and IPV, a multicourse bOPV routine with IPV can provide high levels of protection for types 1 and 3 and moderate protection against type 2.[37]

1.4 Vaccine-associated risks

Wildtype poliovirus actively recombines during replication *in vivo* and OPV genomes are likewise susceptible to recombination.[38-40] This rapid recombination allows for precise poliovirus surveillance of nucleotide substitutions over time.[41,42] Thus, poliovirus isolates are organized into three categories: 1) wild poliovirus, which differ from Sabin strains at >15% of VP1 nucleotides[43]; 2) OPV-like isolates, which vary from Sabin strains by <1% of VP1 nucleotides; and 3) vaccine-derived polioviruses (VDPV) which have ≥10 nucleotide substitutions and differ from Sabin strains by 1-15% of VP1 nucleotides.[7]

The attenuated serotypes used in OPV are produced from a point mutation in the internal ribosome entry site. Sabin strains for types 2 and 3 are attenuated via a few base differences, more for type 1. These attenuating mutations are unstable and Sabin strains are susceptible to reverting towards a wild-type virulent strain.[3] Given the gut environment, Sabin strains tend to

mutate within two to five weeks among 50-100% of vaccinated individuals. Often this mutation is a genetic change from the Sabin strain towards a wild-type genome, in rare cases OPV can regain wild-type virulence.[8] When this happens, OPV can cause vaccine-associated paralytic polio (VAPP) or allow for transmission of VDPV.[44] VAPP is quite rare, estimated to occur in 1 per 2.4 million doses of vaccine administered. VAPP primarily affects the vaccinated individual, and though paralysis has been observed in a close contact, VAPP has not been associated with any outbreak.[2] VDPV, however, is the biggest risk surrounding the poliovirus vaccines and occurs when a vaccine strain of poliovirus undergoes genetic changes in the intestine to revert to wild-type virulence and transmissibility. While recombination is spurious, revertant strains are selected during replication in the gut. Spread of VDPV is encouraged by viral transmissibility dynamics, population immunity levels, sanitation, hygiene behaviors, and the efficiency and effectiveness of control methods.[45] Population immunity, population displacement, diarrheal disease, and high birth rate are indicators of VDPV risk.[46-48] As circulating WPV continues to lessen, VDPV represents the current threat of poliovirus morbidity and the remaining challenge for poliovirus eradication. Accurate estimates of VDPV transmission risk have been difficult to define yet are an important element to end stage polio eradication. A 1999 review of the literature identified OPV2 as more robust, more often excreted, and generally more transmissible than OPV1 or -3.[8] This may help explain the current state of cVDPV2 outbreaks. Currently, the WHO has classified outbreaks of cVDPV2 as a Public Health Emergency of International Concern[49].

1.5 Poliomyelitis Detection and Diagnosis

Surveillance of poliovirus is difficult as a majority of cases are asymptomatic and symptomatic cases most often present with nonspecific symptoms that don’t result in a

healthcare visit. Poliovirus surveillance is largely done via the syndromic surveillance of AFP. The detection of poliovirus from the stool of an AFP case is the gold standard for poliovirus surveillance.[50] Following a clinically confirmed case of AFP, two stool specimens should be collected within 14 days after onset of paralysis, 24-48 hours apart and pathogen analysis should be performs by a qualified laboratory.[13,27,51] Specimen testing is a critical element of polio surveillance as AFP is a symptom of other diseases, most notably Guillain-Barré syndrome, and other non-polio enteroviruses. However, while surveillance for AFP is logistically feasible, it is neither highly sensitive nor specific for detecting poliovirus.[13,52] As AFP occurs in only ~1% of poliovirus infections, surveillance on polioviruses using AFP likely underestimate the true incidence of poliovirus by a factor of 100. Environmental surveillance of poliovirus is also practiced and recommended in areas where AFP surveillance may be deficient or the population is at high risk for viral circulation. As infected individuals can excrete poliovirus for up to several weeks, environmental surveillance of wastewater can provide estimates of size and duration of circulating virus.[50]

Poliovirus serotypes are identified by their major capsid protein, VP1 (900-906 nucleotides).[44] Among each serotype, poliovirus isolates are further categorized as wild-type or vaccine-related. Wild-type polio viruses have no evidence of genetic derivation from a vaccine strain and have demonstrated ability of person-to-person transmission.[44] Vaccine-related polioviruses are classified in one of two ways: OPV-like isolates, which have close genetic relation to one of the developed Sabin strains, or vaccine-derived polioviruses (VDPVs). VDPV strains have undergone sustained transmission and genetic recombination and are identified via their VP1 divergence from the Sabin strains.[44] VDPVs are classified as circulating (cVDPV) when there is demonstrated viral transmission within a community. In addition to cVDPV,

iVDPV describes VDPVs from individuals with immunodeficiencies, and ambiguous VDPVs (aVDPV) describe isolates of an unknown origin.[2,44,45] Natural molecular change for poliovirus is around a 1% nucleotide substitution per year.[45,53,54] Thus, isolates that differ from OPV-like strains by >1% nucleotide positions are thought to have replicated for at least a year in one or more hosts and are considered cVDPVs. However, since poliovirus type 2 is responsible for 87-90% of cVDPVs, to increase surveillance sensitivity of cVDPV2 outbreaks, the cut off for VDPV designation was lowered to 0.6% nucleotide substitution for poliovirus type 2.[2,44] Laboratory characterization of isolates are done via real-time reverse transcription polymerase chain reaction (rRT-PCR). In the current climate of the polio endgame strategy, circulation of VDPVs represent the greatest polio-related health risk as they carry the potential to become endemic in areas of low vaccination coverage. The Global Polio Laboratory Network (GPLN) was established in 1990 by the WHO and participating nations. It is a network of 146 facilities in 92 countries working to distinguish poliovirus as the causative agent of AFP from patient specimens and in select laboratories, from sewage samples.[55,56] The GPLN follows standardized protocols to identify WPV, screen for OPV-like isolates or VDPVs, and conduct genomic sequencing.

1.6 Poliovirus Prevalence & Incidence

Prior to the development of the poliovirus vaccines (pre-1955), nearly all children were infected and around 1 in 200 individuals developed paralytic poliomyelitis.[2,20] WPV1 accounted for nearly 80% of paralytic cases. In the prevaccine era, poliomyelitis followed seasonal trends in temperate zones—epidemic peaks were generally between August-October. Across the globe poliovirus followed a latitudinal gradient and tended to break away from seasonal trends towards the equator.[20,57] Some hypothesize that seasonality was one of the factors that allowed temperate

regions, particularly the global North, to effectively plan and eradicate poliomyelitis.[12] The sustained use of polio vaccines globally has been one of the most successful public health programs ever conducted on a global scale.[12]

In 1988, there was an estimated 350,000 cases of poliomyelitis in 125 countries.[58] Since then, WPV incidence has dropped more than 99%. The last case of WPV2 was observed in 1999 in Uttar Pratesh, India and in 2015 the WHO officially declared WPV2 eradicated.[53,59] WPV3 was last observed on November 10, 2012 in an 11 year old boy in Yoba, northern Nigeria and was declared eradicated in October 2019.[60] Following and verifying the eradication of WPV has been possible due to the uniqueness of nucleotide sequences in different geographic regions.[12] Since the final WPV3 case in 2012, only WPV1 has been detected in just three countries: Nigeria[61-63], Afghanistan, and Pakistan.[1] Persistence of WPV transmission can be attributed to the failure of vaccination campaigns, failure of the vaccine to cause seroconversion, and the viral epidemiology.

Nigeria was considered free of endemic WPV1 in September, 2015 but almost a year later in August, 2016 WPV1 was detected in Borno State.[64] Genetic sequencing of the outbreak strain suggests transmission has been ongoing but undetected since 2013.[65] While a WPV1 case has not been observed since 2016, in Borno State violent conflict due to the insurgency of Boko Haram has limited vaccination efforts creating risk of both WPV and VDPV emergence.[63] It is estimated that 130,000-210,000 (28-45%) of the 469,000 eligible children in these inaccessible areas are unvaccinated. A national study conducted in 2016-2017 found that overall 33% of children were fully vaccinated against poliovirus but by region this figure ranged widely from 7-75%. Median coverage across all districts in Nigeria was below 50%.[61,66]

The persistence of WPV1 in Pakistan and Afghanistan is similarly tied to disruption of the healthcare system due to armed conflict. A recent study found that around 20% of children were inaccessible to vaccinators and that in areas of high insecurity, vaccination rates were 5.3% lower than in secure areas. Additionally, polio incidence increased by 73% during times of high insecurity.[67] Endemic transmission in Pakistan and Afghanistan continues, but incidence rate are at a historic low.[46] In 2017, 22 cases of WPV1 were detected in this region and 18 cases from January- September 2018.[63]

In the Democratic Republic of the Congo (DRC), periods of conflict in the east has posed a threat to polio eradication efforts. Owing to this, the DRC was the last country in Africa to implement national immunization days in 1999. Since then, subsequent mass vaccination activities have been disrupted due to violence and hostile attitudes towards vaccination teams.[68] Since 2000, cVDPV outbreaks have occurred in 18 countries. Type 2 associated VDPV represents the overwhelming majority of cases, 11.1% of cases are associated with type 1, and just 1.8% of reported cases are cVDPV3. Looking at cases just since 2005, type 2 represents 97.1% of all cVDPV infections.[44] Most recently from January 2017-June 2018, six countries detected cVDPV outbreaks via AFP surveillance, non-AFP polio cases, and environmental surveillance. DRC, Kenya, Nigeria, Somalia, and Syria reported cVDPV2 circulation and Papua New Guinea reported cVDPV1 (Table 1.1).

Global Eradication Efforts and The Strategic Plan

Albert Sabin advocated for mass OPV campaigns as the most effective control method for polio. In 1962, Cuba was the first to initiate mass campaigns and stopped WPV circulation within a year. The implementation and success of national immunization days (NIDs) in Brazil and Mexico in the early 1980s led the Pan American Health Organization (PAHO) to

recommend NIDs to all member states and resolved to eradicate polio by 1990s from the Americas.[7,8] Following PAHO's achievements, in 1988 the World Health Assembly resolved to eradicate poliomyelitis by the year 2000 and launched the Global Polio Eradication Initiative (GPEI) to facilitate this.[13,58,65] In areas where eradication has yet to be declared, the strategy of national level Polio Eradication Initiatives is to foster high routine immunization (RI) coverage for children under 5, supplement RI with NIDs, target supplementary immunization campaigns (SIAs) for village based "mop up" immunization campaigns, and ensure sensitive poliovirus surveillance is maintained.[7]

Despite the achievement of WPV2 eradication VDPV events have been increasing since the early 2000s, motivating the WHO to reassess poliovirus control logistics.[44] In May 2012 the World Health Assembly called for the development of a comprehensive polio endgame strategy. Endorsed by World Health Organization (WHO) Member States in 2013, *The Polio Eradication and Endgame Strategic Plan 2013-2018* (heretofore referred to as *The Strategic Plan*) had four main objectives:

1. Detect and interrupt poliovirus transmission
2. Strengthen immunization programmes, introduce at least one dose of inactivated poliomyelitis vaccine (IPV) and withdraw oral poliomyelitis vaccine (OPV), starting with type 2 component
3. Contain poliovirus and certify the interruption of transmission
4. Plan for the legacy of the polio endgame

To achieve these aims, the WHO identified the need to ultimately end OPV use due to the risks of VDPVs associated with the vaccine. To initiate this process, removing poliovirus type 2 from the OPV was identified as a first step. The continued use of a tOPV was contributing to the

emergence of cVDPV, 90% of which are identified at type 2. In April 2016, the WHO facilitated a global "switch" day in which global stocks of tOPV were replaced with a bivalent OPV (contains type 1 and 3).[2] By May 12, 2016 all 155 countries and territories that used OPV in 2015 has switched to bOPV.[69] While removal of tOPV facilitated the long-term eradication goals, due to the policy change, the population immunity to type 2 will wane and new birth cohorts will be susceptible to poliomyelitis while cohorts previously immunized with tOPV remain in the community.[70,71] To mitigate this risk, *The Strategic Plan* recommended that countries which still administer OPV add one dose of IPV at 14 weeks of age to reduce the risk of sustained transmission of poliovirus type 2.[69,72] In polio-endemic countries an additional birth dose of bOPV was added to maximize seroconversion and induce mucosal protection (Table 1.2).[1,44] To ensure that the switch indeed occurred in health centers across the globe and that inadvertent use of tOPV does not occur, internal and external monitors visited over 160,000 health centers and vaccine stores to verify the switch.[69] As a further precaution, around 100 million doses of mOPV2 were distributed to 11 countries and are maintained for use against WPV2 or more likely, VDPV2 circulation.[63] Use for any reason must be authorized by the director general with guidance from an independent advisory group.[73]

Today, circulation of VDPV2 in 24 countries has problematized current polio vaccine strategies. *The Strategic Plan* was predicated on the assumption that cessation of Sabin-strain polio vaccines would be possible. However, the transmissibility of OPV has complicated the phased withdrawal plan.[74] To avoid a potential need to restart tOPV vaccination, which could seed new cVDPV2 outbreaks, a novel oral polio vaccine (nOPV2) has been developed for special use in VDPV2-endemic countries.[75,76] nOPV2 has been engineered to lower the risk of reversion to neurovirulence and is intended to be used in lieu of mOPV2.[77-79]

1.7 Poliovirus in the DRC

In DRC the Extended Program on Immunization (EPI) was introduced in 1978. In 1996, EPI created the Polio Eradication Program which began providing additional doses of OPV through SIAs in areas with high disease burden.[60] In 1999 NIDs were initiated but the campaigns lasted only through 2002 until 2011 when they were resumed.[80]

Polio was considered endemic until 2001, when there was an interruption of transmission and no WPV cases reported for four years.[80] However, from 2006-2011 importations of WPV from neighboring Angola caused sustained WPV1 and WPV3 outbreaks. From 2006-2009 DRC saw 58 cases of WPV1 and 4 cases of WPV3 all of which were in children less than 15 years of age. However, from 2010-2011 the outbreak escalated. In 2010 there were 100 WPV1 cases in 5 provinces (Bandundu, Bas Congo, Kasaï Occidental, Katanga, Kinshasa) and in 2011, 93 cases in six provinces (addition of Maniema). An average of 17% (range: 0-50%) of these cases occurred in individuals 15 years or older.[80] Prior to this outbreak all observed WPV cases had been in children <15 years of age. DRC responded to this outbreak by conducting SIAs from 2010-2012 with mOPV1, bOPV, and tOPV. However, these campaigns were targeted towards children aged <5 years. An analysis by Alleman suggests that this WPV1 outbreak could be explained by an immunity gap in poliovirus serotype 1 due to suboptimal exposure to OPV. The last confirmed WPV1 infection was in Maniema province on December 20, 2011.[44,60,81] The last reported case of WPV2 was documented in 1997.

The DRC has experienced cases of documented VDPVs since 2004.[81,82] In 2011-2012 Haut Lomami province was the source of an outbreak of 30 VDPV cases, the only reported VDPV cases from that time. After this outbreak through 2016, no outbreaks of VDPV were reported and only five isolated VDPV cases were detected nationally. However, in 2017 VDPV

reemerged in Haut Lomami and the neighboring Tanganyika and Maniema provinces. Of the 25 VDPV cases reported from 2017, in Haut Lomami 20 were considered a cVDPV and two were considered ambiguous—their origins could not be traced to an earlier case—two VDPV cases were linked in Maniema, and one aVDPV case was recorded in Tanganyika. In 2018, 22 cases of VDPV were reported from five provinces. Of these, 11 cVDPV cases were reported from Mongala province in the northwest of the country. In the southeast, Haut Lomami reported two cases of cVDPV, Tanganyika reported two cases of cVDPV and one case of aVDPV, and Haut Katanga, a neighbor of the two reported four cases of cVPDV. (Figure 1.2) These case reports speak to a larger trend in the DRC. Since 2011 there has been continuing cases of VDPV in Haut Lomami and Tanganyika that raise questions about the vaccination landscape of this particular region. However, the dramatic increase in 2018 in case reports from Mongala province in the northwest, speaks to a broader nationwide concern of low poliovirus vaccination rates that perhaps is just being evidenced now. In 2019, there were 68 reported cVDPV2 cases from nine health zones. Of these Sankuru reported 21 and Haut Lomami reported 18.

On February 13, 2018 the DRC Ministry of health declared cVDPV2 to be a national public health emergency.[83] In October, 2018 the DRC announced a new plan to increase routine immunizations across the nation. The Emergency Plan for the Revitalization of Routine Immunization in the DRC, nicknamed Plan Mashako after former Minister of Health Leonard Mashako Mamba was spearheaded by GAVI and its implementation is led by EPI with support from the DRC ministry of health, WHO, and UNICEF. Plan Mashako is focused on nine priority provinces (Ituri, Kasai, Upper Katanga, Mongala, Kwilu, Tanganyika, Kinshasa, Tshuapa and Haut-Lomami) and aims to increase routine immunization rates by 15% by vaccinating 200,000

additional children from October 2018- April 2020.[84] In order to achieve this Plan Mashako activities are grouped into 5 themes (Table 1.3).

To measure progress, indicators are measures and reported monthly: availability of vaccines, health areas respecting the number of immunization sessions required, health zone supervision activities by provincial health division supervisors, supervision activities of health areas by health zone supervisors, and cold chain functionality. To support Plan Mashako and increase routine immunization, the DRC Ministry of Health opened the Kinkole immunization hub outside of Kinshasa, one of the continent's largest vaccine storage facilities with around 6,000m^3 of storage space.

1.8 Gaps in the Literature & Project Impact

In the DRC the last confirmed WPV case was recorded in Maniema province in December 20, 2011. Since then however there has been consistent spread of cVPDVs in the eastern provinces of Maniema, Haut Lomami, and Tanganyika (Figure 1.2). Additionally, AFP case counts have increased from 1820 to 2148 to 2751 from 2016-2018, respectively. Due to the ~1% AFP rate of poliovirus and the poor viral surveillance of this region, these data indicate the likelihood of a larger poliovirus outbreak than current surveillance estimates. (Rate of non-polio AFP estimated at 3.3-5.2%).

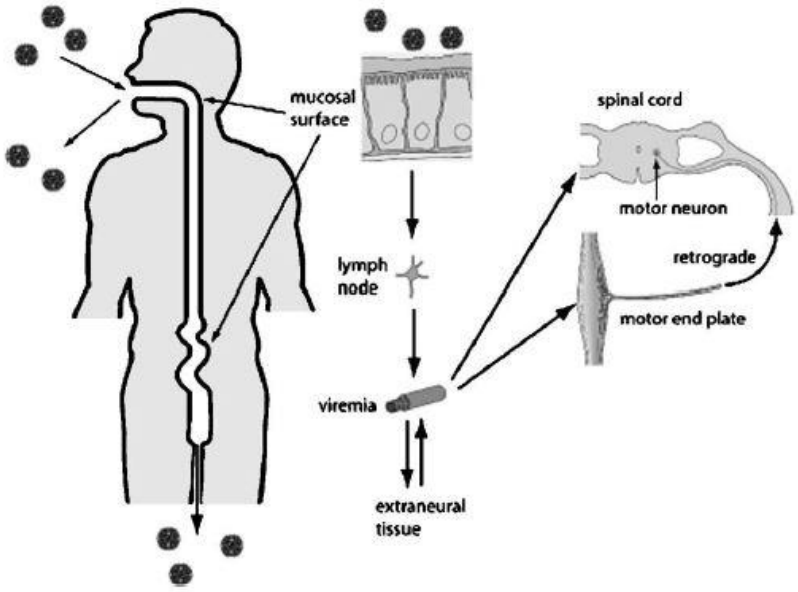
As cVDPV2 cases continue to increase they threaten the polio eradication plan which aims to eventually remove OPV from routine vaccination efforts. Should cVDPV2 continue to increase in DRC OPV2 would need to be reintroduced, stalling eradication efforts. To prevent the emergence and spread of VDPVs, high vaccine coverage is paramount. However, there is a limited current understanding about where this threshold lies, and the current state of polio seroprevalence in the DRC. Additionally, in response to observed VDPV2 cases the DRC uses

SIAs to immunize at-risk areas. While this strategy is in line with the recommendations of GPEI, its effectiveness in logistically challenging outbreak settings like the DRC has not been confirmed. In the DRC the consistent incidence of cVDPV2 cases despite frequent SIA campaigns which use mOPV2 may point to a failure in the SIA delivery pipeline. The efficacy of SIAs and the ways in which they can be improved have not yet been analyzed in the DRC.

Our research group has conducted serologic and demographic surveys of the population in Haut Lomami and Tanganyika provinces since 2014 to better understand the current status of seroimmunity prevalence and any demographic factors or knowledge and attitudes that may contribute towards low vaccination rates and continued cVDPV transmission.

1.9 Tables and Figures

Figure 1.1. Schematic of poliovirus pathogenesis



From V.R. Racaniello, 2016[3]

Table 1.1. Global Circulating cVDPVs

From: http://polioeradication.org/polio-today/polio-now/this-week/circulating-vaccine-derived-poliovirus/

World Health Organization

Global Circulating Vaccine-derived Poliovirus (cVDPV)[1,2,3]

Country	AFP cases (Paralysis onset between 2000-2019)						Other sources (Human)[5] (collection between 2015-2019)						Other sources (Environment) (collection between 2015-2019)					
	cVDPV1																	
	2015	2016	2017	2018	2019	Onset of most recent	2015	2016	2017	2018	2019	most recent collection	2015	2016	2017	2018	2019	most recent collection date
Philippines					1	28-Oct-19											14	28-Nov-19
Malaysia					1	26-Oct-19											1	16-Nov-19
Myanmar					6	09-Aug-19					6	21-Aug-19						
Indonesia				1		27-Nov-18					2	13-Feb-19						
PNG				26		18-Oct-18				7		20-Sep-18				7		06-Nov-18
Laos	8	3				11-Jan-16	6	5				09-Feb-16						
Madagascar	10					22-Aug-15	1					01-Aug-15						
Ukraine	2					07-Jul-15												
Total type 1	20	3	0	27	8		7	5	0	7	8		0	0	0	7	15	
	cVDPV2																	
Country	2015	2016	2017	2018	2019	Onset of most recent	2015	2016	2017	2018	2019	most recent collection	2015	2016	2017	2018	2019	most recent collection date
Pakistan	2	1			12	22-Nov-19					20	10-Oct-19	7	4			25	04-Dec-19
Central African Republic					17	09-Nov-19					38	30-Nov-19					9	25-Sep-19
Zambia					2	25-Nov-19					2	25-Sep-19						
Philippines					10	25-Oct-19					6	23-Nov-19					17	06-Nov-19
Ghana					11	07-Nov-19					12	23-Nov-19					11	20-Nov-19
Malaysia																	1	16-Nov-19
Angola					86	04-Nov-19					21	28-Oct-19					11	11-Nov-19
Nigeria	1	1		34	18	09-Oct-19		2[2]		53	18	24-Jul-19	2	1		44	58	11-Nov-19
Somalia				6[6]	3	08-May-19				3	2	25-May-19			2	19	2	10-Nov-19
Ethiopia					5	09-Sep-19					8	06-Nov-19					1	21-Oct-19
DR Congo			22	20	63	31-Oct-19			19	15	18	21-Oct-19						
Togo					4	28-Oct-19												
Chad					3	28-Oct-19					2	10-Oct-19						
Benin					6	15-Oct-19												
Côte d'Ivoire																	2	26-Sep-19
China					1	25-Apr-19					3	18-Aug-19				1		18-Apr-18
Cameroon																	1	20-Apr-19
Niger				10	1	03-Apr-19				4	6	16-Mar-19						
Mozambique				1		21-Oct-18				2		17-Dec-18						
Kenya						29-Aug-12										1		21-Mar-18
Syria			74			21-Sep-17		1[4]	66			12-Sep-17						
Guinea	7					14-Dec-15												
Myanmar	2					05-Oct-15												
Total type 2	12	2	96	71	242		0	3	85	77	156		9	5	2	65	138	
	cVDPV3																	
Country	2015	2016	2017	2018	2019	Onset of most recent	2015	2016	2017	2018	2019	most recent collection	2015	2016	2017	2018	2019	most recent collection date
Somalia				7[6]		07-Sep-18				2		29-Jun-18				11		23-Aug-18
Total type 3	0	0	0	7	0		0	0	0	2	0		0	0	0	11	0	

Environmental surveillance for poliovirus in selected sewage sites established and working

Changes from previous week

[1]For cVDPV definition see http://polioeradication.org/wp-content/uploads/2016/09/Reporting-and-Classification-of-VDPVs_Aug2016_EN.pdf . Niger 2006, Niger 2009, Niger 2010, Chad 2010 cVDPVs are linked to the Nigeria outbreak. Kenya 2012 cVDPVs are linked to the Somalia outbreak. Nigeria figures include cases with WPV1/cVDPV2 mixture: 2005 - 2, 2006 - 1, 2007 - 1, 2008 - 3, 2009 - 1, 2011 - 1; WPV3/cVPDV2 mixture: 2007 - 2. [2] include a cVDPV2 from a contact of a WPV1 case in Nigeria. [3]Figures include multiple emergences. [4] stool collected in Sep - 2016 but the final result was reported in 2017. [5] Include contact, healthy and community samples . Positive contact of a negative index AFP case double counted in both AFP cases and other sources count . [6] 1 cVDPV2 and cVDPV3 isolated from one child

Data in WHO HQ as of 07 Jan. 2020

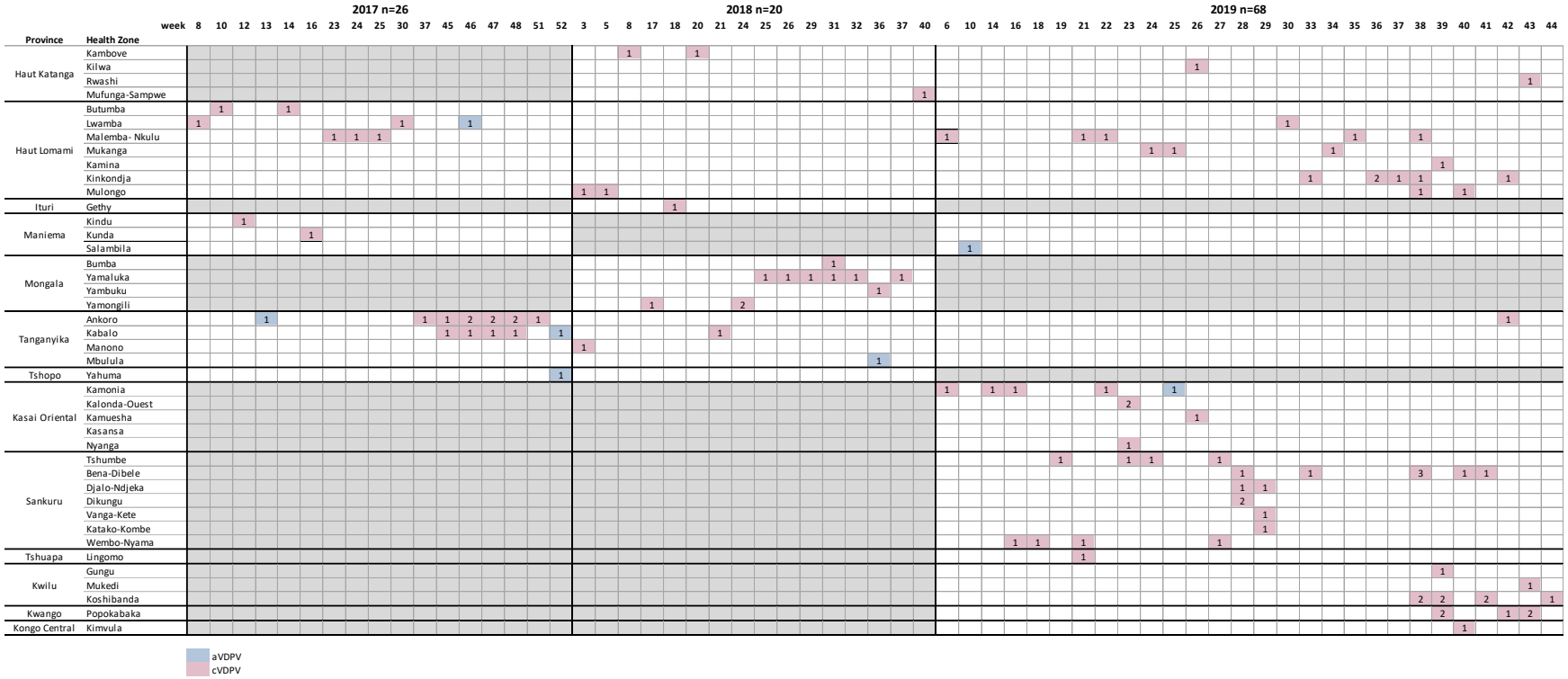
Table 1.2. Current Polio Vaccination Schedule in DRC [85]

Time	Vaccine
Birth	OPV
6 weeks	OPV
10 weeks	OPV
14 weeks	OPV + IPV

Table 1.3. Plan Mashako thematic activities

Themes	Objectives	Innovations
Coordination and steering	Put in place committees to ensure speedy implementation at all levels	Weekly meetings of the steering committee at the levels national and provincial level to ensure rapid implementation. Half-yearly report with the Minister of Health.
Provision of services and communication	Increase the number of immunization sessions by at least 20% Reduce abduction rate to less than 10%	Simple Vaccination Planning Tool Setting Targets for Each Health Zone, Health Zone, and Province. Creation of CACs and support of the community dynamic
Vaccine availability	Reduce local stock-outs by 80%	Planning in advance of deliveries to provinces. Delivery system adapted to the country's logistics challenges. Systematic inventory tracking at all levels
Monitoring and evaluation	Monitor monthly progress in each health zone, province and at the national level	Dashboard providing real-time data to act quickly. Monthly reports sent to health zones and provinces
Inspection and control	Inspect immunization activities each month in each health zone	Systematic control of immunization activities to verify and validate performance

Figure 1.2. Laboratory-Confirmed VDPV2 cases by Heath Zone and Week, 2017-2019, DRC



1.10 References

1. World Health Organization. Polio vaccines: WHO position paper, March 2016– recommendations. *Vaccine.* 2017;35(9):1197-1199.

2. World Health Organization. *A guide to introducing Inactivated Poliomyelitis Vaccine based on the Polio Eradication & Endgame Strategic Plan 2013-2018.* 2016.

3. Racaniello VR. One hundred years of poliovirus pathogenesis. *Virology.* 2006;344(1):9-16.

4. Racaniello VR. Picornaviridae, the virus and their replication. *Fields virology.* 2007:795-838.

5. Blondel B, Duncan G, Couderc T, Delpeyroux F, Pavio N, Colbàre-Garapin F. Molecular aspects of poliovirus biology with a special focus on the interactions with nerve cells. *Journal of neurovirology.* 1998;4(1):1-26.

6. Bodian D. Differentiation of Types of Poliomyelitis Viruses. I. Reinfection Experiments in Monkeys (Second Attacks). *American journal of hygiene.* 1949;49(2):200-218.

7. Kew OM, Sutter RW, Gourville EMd, Dowdle WR, Pallansch MA. Vaccine-Derived Polioviruses and the Endgame Strategy for Global Polio Eradication. *Annual Review of Microbiology.* 2005;59(1):587-635.

8. Fine PEM, Carneiro IAM. Transmissibility and Persistence of Oral Polio Vaccine Viruses: Implications for the Global Poliomyelitis Eradication Initiative. *American Journal of Epidemiology.* 1999;150(10):1001-1021.

9. Modlin JF. Poliomyelitis and Poliovirus Immunization. In: *Human Enterovirus Infections.* American Society of Microbiology; 1995.

10. Helleringer S, Abdelwahab J, Vandenent M. Polio supplementary immunization activities and equity in access to vaccination: evidence from the demographic and health surveys. *The Journal of infectious diseases.* 2014;210(suppl_1):S531-S539.

11. Centers for Disease Control and Prevention. *Epidemiology and prevention of vaccine-preventable diseases.* 13th ed. Washington DC: Public Health Foundation; 2015.

12. Nathanson N, Kew OM. From Emergence to Eradication: The Epidemiology of Poliomyelitis Deconstructed. *American Journal of Epidemiology.* 2010;172(11):1213-1229.

13. Marx A, Glass JD, Sutter RW. Differential Diagnosis of Acute Flaccid Paralysis and its Role in Poliomyelitis Surveillance. *Epidemiologic Reviews.* 2000;22(2):298-316.

14. Sartwell PE. The Incubation Period of Poliomyelitis. *American Journal of Public Health and the Nations Health.* 1952;42(11):1403-1408.

15. Horstmann DM, Paul JR. The incubation period in human poliomyelitis and its implications. *Journal of the American Medical Association.* 1947;135(1):11-14.

16. Chapter 25 - Vaccine-Preventable Diseases. In: Flaherty DK, ed. *Immunology for Pharmacy.* Saint Louis: Mosby; 2012:197-213.

17. Topley WWC, Wilson GS, Collier LH, et al., eds. *Topley & Wilson's microbiology and microbial infections.* 9th ed. London; New York: Hodder Arnold ; ASM Press; 2005. Virology; No. vol 1.

18. World Health Organization. The Immunological Basis for Immunization Series: Module 6: Poliomyelitis. In:1996.

19. Routh JA, Oberste MS, Patel M. Manual for the surveillance of vaccine-preventable diseases. Chapter 12: Poliomyelitis. 2012.

20. Nathanson N, Martin JR. The Epidemiology of Poliomyelitis: Enigmas Surrounding its Appearance, Epidemicity, and Disappearance. *American Journal of Epidemiology.* 1979;110(6):672-692.

21. Sabin AB. Oral Poliovirus Vaccine: History of Its Development and Use and Current Challenge to Eliminate Poliomyelitis from the World. *The Journal of Infectious Diseases.* 1985;151(3):420-436.

22. Kabir M, Afzal MS. Epidemiology of polio virus infection in Pakistan and possible risk factors for its transmission. *Asian Pacific Journal of Tropical Medicine.* 2016;9(11):1044-1047.

23. Minor PD. Poliovirus biology. *Structure.* 1996;4(7):775-778.

24. Minor P. Antigenic structure of picornaviruses. *Picornaviruses.* 1990:121-154.

25. Gromeier M, Nomoto A. Determinants of poliovirus pathogenesis. *Molecular biology of picornavirus.* 2002:367-379.

26. Hogle JM. Poliovirus cell entry: common structural themes in viral cell entry pathways. *Annual Reviews in Microbiology.* 2002;56(1):677-702.

27. Alexander Jr JP, Gary Jr HE, Pallansch MA. Duration of poliovirus excretion and its implications for acute flaccid paralysis surveillance: a review of the literature. *The Journal of infectious diseases.* 1997;175(Supplement_1):S176-S182.

28. Nathanson N. Chapter 1 The Pathogenesis of Poliomyelitis: What We Don't Know. In: *Advances in Virus Research.* Vol 71. Academic Press; 2008:1-50.

29. Shahzad A, Köhler G. Inactivated Polio Vaccine (IPV): A strong candidate vaccine for achieving global polio eradication program. *Vaccine.* 2009;27(39):5293-5294.

30. Parker EP, Molodecky NA, Pons-Salort M, O'Reilly KM, Grassly NC. Impact of inactivated poliovirus vaccine on mucosal immunity: implications for the polio eradication endgame. *Expert review of vaccines.* 2015;14(8):1113-1123.

31. The Cuba IPV Study Collaborative Group. Randomized, Placebo-Controlled Trial of Inactivated Poliovirus Vaccine in Cuba. *New England Journal of Medicine.* 2007;356(15):1536-1544.

32. Henry JL, Jaikaran ES, Davies JR, et al. A study of poliovaccination in infancy: excretion following challenge with live virus by children given killed or living poliovaccine. *J Hyg (Lond).* 1966;64(1):105-120.

33. Laassri M, Lottenbach K, Belshe R, et al. Effect of Different Vaccination Schedules on Excretion of Oral Poliovirus Vaccine Strains. *The Journal of Infectious Diseases.* 2005;192(12):2092-2098.

34. Famulare M, Chang S, Iber J, et al. Sabin Vaccine Reversion in the Field: a Comprehensive Analysis of Sabin-Like Poliovirus Isolates in Nigeria. *Journal of Virology.* 2016;90(1):317-331.

35. Sutter RW, John TJ, Jain H, et al. Immunogenicity of bivalent types 1 and 3 oral poliovirus vaccine: a randomised, double-blind, controlled trial. *The Lancet.* 2010;376(9753):1682-1688.

36. Mateen FJ, Shinohara RT, Sutter RW. Oral and inactivated poliovirus vaccines in the newborn: a review. *Vaccine.* 2013;31(21):2517-2524.

37. Bandyopadhyay AS, Modlin JF, Wenger J, Gast C. Immunogenicity of New Primary Immunization Schedules With Inactivated Poliovirus Vaccine and Bivalent Oral Polio Vaccine for the Polio Endgame: A Review. *Clinical infectious diseases : an official publication of the Infectious Diseases Society of America.* 2018;67(suppl_1):S35-S41.

38. Kew OM, Nottay BK. Molecular epidemiology of polioviruses. *Reviews of infectious diseases.* 1984;6 Suppl 2:S499-504.

39. Minor PD, Ferguson M, Evans DM, Almond JW, Icenogle JP. Antigenic structure of polioviruses of serotypes 1, 2 and 3. *The Journal of general virology.* 1986;67 (Pt 7):1283-1291.

40. Cuervo NS, Guillot S, Romanenkova N, et al. Genomic Features of Intertypic Recombinant Sabin Poliovirus Strains Excreted by Primary Vaccinees. *Journal of Virology.* 2001;75(13):5740-5751.

41. Liu H-M, Zheng D-P, Zhang L-B, Oberste MS, Pallansch MA, Kew OM. Molecular evolution of a type 1 wild-vaccine poliovirus recombinant during widespread circulation in China. *Journal of Virology.* 2000;74(23):11153-11161.

42. Shulman LM, Handsher R, Yang C-F, et al. Resolution of the pathways of poliovirus type 1 transmission during an outbreak. *Journal of clinical microbiology.* 2000;38(3):945-952.

43. WHO. Expanding contributions of the Global Laboratory Network for Poliomyelitis Eradication. *Wkly Epidemiol Rec.* 2002;77:133-137.

44. Burns CC, Diop OM, Sutter RW, Kew OM. Vaccine-Derived Polioviruses. *The Journal of Infectious Diseases.* 2014;210(suppl_1):S283-S293.

45. Wringe A, Fine PEM, Sutter RW, Kew OM. Estimating the Extent of Vaccine-Derived Poliovirus Infection. *PLOS ONE.* 2008;3(10):e3433.

46. O'Reilly KM, Lamoureux C, Molodecky NA, Lyons H, Grassly NC, Tallis G. An assessment of the geographical risks of wild and vaccine-derived poliomyelitis outbreaks in Africa and Asia. *BMC Infectious Diseases.* 2017;17:367.

47. Wassilak S, Pate MA, Wannemuehler K, et al. Outbreak of type 2 vaccine-derived poliovirus in Nigeria: emergence and widespread circulation in an underimmunized population. *J Infect Dis.* 2011;203(7):898-909.

48. Burns CC, Shaw J, Jorba J, et al. Multiple Independent Emergences of Type 2 Vaccine-Derived Polioviruses during a Large Outbreak in Northern Nigeria. *Journal of Virology.* 2013;87(9):4907.

49. Zarocostas J. WHO keeps polio on the international health emergency list. *The Lancet.* 2018;392(10163):2425.

50. World Health Organization. Guidelines for environmental surveillance of poliovirus circulation. 2003.

51. Levitt A, Diop OM, Tangermann RH, et al. Surveillance Systems to Track Progress Toward Global Polio Eradication — Worldwide, 2012–2013. *MMWR Morbidity and Mortality Weekly Report.* 2014;63(16):356-361.

52. Kew OM, Mulders MN, Lipskaya GY, da Silva EE, Patlansch MA. Molecular epidemiology of polioviruses. *Seminars in Virology.* 1995;6(6):401-414.

53. Jorba J, Campagnoli R, De L, Kew O. Calibration of Multiple Poliovirus Molecular Clocks Covering an Extended Evolutionary Range. *Journal of Virology.* 2008;82(9):4429-4440.

54. Gavrilin GV, Cherkasova EA, Lipskaya GY, Kew OM, Agol VI. Evolution of Circulating Wild Poliovirus and of Vaccine-Derived Poliovirus in an Immunodeficient Patient: a Unifying Model. *Journal of Virology.* 2000;74(16):7381-7390.

55. Global Polio Eradication Initiative. The Global Polio Laboratory Network. 2018; http://polioeradication.org/polio-today/polio-now/surveillance-indicators/the-global-polio-laboratory-network-gpln/. Accessed 10/13/2018.

56. Centers for Disease Control and Prevention. Laboratory surveillance for wild and vaccine-derived polioviruses--worldwide, January 2007-June 2008. *MMWR Morbidity and mortality weekly report.* 2008;57(35):967.

57. Martinez-Bakker M, King AA, Rohani P. Unraveling the Transmission Ecology of Polio. *PLoS Biology.* 2015;13(6):e1002172.

58. Khan F, Datta SD, Quddus A, et al. Progress toward polio eradication—worldwide, January 2016–March 2018. *Morbidity and Mortality Weekly Report.* 2018;67(18):524.

59. Centers for Disease Control and Prevention. Apparent global interruption of wild poliovirus type 2 transmission. *MMWR Morbidity and mortality weekly report.* 2001;50(12):222-224.

60. Voorman A, Hoff NA, Doshi RH, et al. Polio immunity and the impact of mass immunization campaigns in the Democratic Republic of the Congo. *Vaccine.* 2017;35(42):5693-5699.

61. Bolu O, Nnadi C, Damisa E, et al. Progress toward poliomyelitis eradication—Nigeria, January–December 2017. *Morbidity and Mortality Weekly Report.* 2018;67(8):253.

62. Giwa FJ, Olayinka AT, Ogunshola FT. Seroprevalence of poliovirus antibodies amongst children in Zaria, Northern Nigeria. *Vaccine.* 2012;30(48):6759-6765.

63. Jorba J, Diop OM, Iber J, et al. Update on vaccine-derived polioviruses—worldwide, January 2017–June 2018. *Morbidity and Mortality Weekly Report.* 2018;67(42):1189.

64. Craig KT, Verma H, Iliyasu Z, et al. Role of Serial Polio Seroprevalence Studies in Guiding Implementation of the Polio Eradication Initiative in Kano, Nigeria: 2011-2014. *The Journal of infectious diseases.* 2016;213 Suppl 3(Suppl 3):S124-S130.

65. Morales M. Progress toward polio eradication—worldwide, 2015–2016. *MMWR Morbidity and mortality weekly report.* 2016;65.

66. Gunnala R, Ogbuanu IU, Adegoke OJ, et al. Routine Vaccination Coverage in Northern Nigeria: Results from 40 District-Level Cluster Surveys, 2014-2015. *PLoS ONE.* 2016;11(12):e0167835.

67. Verma AA, Jimenez MP, Tangermann RH, Subramanian SV, Razak F. Insecurity, polio vaccination rates, and polio incidence in northwest Pakistan. *Proceedings of the National Academy of Sciences.* 2018.

68. Haddison EC, Ngono D, Kouamen GT, Kagina BM. Successful polio supplementary immunisation activities in a security compromised zone – Experiences from the Southwest region of Cameroon. *Vaccine.* 2018.

69. Hampton LM, Farrell M, Ramirez-Gonzalez A, et al. Cessation of trivalent oral poliovirus vaccine and introduction of inactivated poliovirus vaccine—worldwide, 2016. *Morbidity and Mortality Weekly Report.* 2016;65(35):934-938.

70. Dowdle WR, De Gourville E, Kew OM, Pallansch MA, Wood DJ. Polio eradication: the OPV paradox. *Reviews in medical virology.* 2003;13(5):277-291.

71. Kroiss SJ, Famulare M, Lyons H, McCarthy KA, Mercer LD, Chabot-Couture G. Evaluating cessation of the type 2 oral polio vaccine by modeling pre- and post-cessation detection rates. *Vaccine.* 2017;35(42):5674-5681.

72. Hovi T. Inactivated poliovirus vaccine and the final stages of poliovirus eradication. *Vaccine.* 2001;19(17):2268-2272.

73. Global Polio Eradication Initiative. *Polio Eradication and Endgame Strategic Plan 2013–2018.* 2014.

74. Fine PE, Oblapenko G, Sutter RW. Polio control after certification: major issues outstanding. *Bulletin of the World Health Organization.* 2004;82(1):47-52.

75. Duintjer Tebbens RJ, Thompson KM. Polio endgame risks and the possibility of restarting the use of oral poliovirus vaccine. *Expert Review of Vaccines.* 2018;17(8):739-751.

76. Fu R, Altamirano J, Sarnquist CC, Maldonado YA, Andrews JR. Assessing the Risk of Vaccine-derived Outbreaks After Reintroduction of Oral Poliovirus Vaccine in Postcessation Settings. *Clinical infectious diseases : an official publication of the Infectious Diseases Society of America.* 2018;67(suppl_1):S26-S34.

77. Yeh MT, Bujaki E, Dolan PT, et al. Engineering the Live-Attenuated Polio Vaccine to Prevent Reversion to Virulence. *Cell Host & Microbe.* 2020;27(5):736-751.e738.

78. Konopka-Anstadt JL, Campagnoli R, Vincent A, et al. Development of a new oral poliovirus vaccine for the eradication end game using codon deoptimization. *npj Vaccines.* 2020;5(1):26.

79. Alleman MM, Jorba J, Greene SA, et al. Update on vaccine-derived poliovirus outbreaks—worldwide, July 2019–February 2020. *Morbidity and Mortality Weekly Report.* 2020;69(16):489.

80. Alleman MM, Wannemuehler KA, Weldon WC, et al. Factors Contributing to Outbreaks of Wild Poliovirus Type 1 Infection Involving Persons Aged ≥15 Years in the Democratic Republic of the Congo, 2010–2011, Informed by a Pre-Outbreak Poliovirus Immunity Assessment. *The Journal of Infectious Diseases.* 2014;210(suppl_1):S62-S73.

81. Alleman MM, Chitale R, Burns CC, et al. Vaccine-Derived Poliovirus Outbreaks and Events - Three Provinces, Democratic Republic of the Congo, 2017. *MMWR Morbidity and mortality weekly report.* 2018;67(10):300-305.

82. Chard AN, Datta SD, Tallis G, et al. Progress Toward Polio Eradication - Worldwide, January 2018-March 2020. *MMWR Morbidity and mortality weekly report.* 2020;69(25):784-789.

83. World Health Organization. Poliomyelitis - Democratic Republic of the Congo. *Disease outbreak news* 2018; https://www.who.int/csr/don/20-December-2018-poliovirus-drc/en/. Accessed 2/27/2020, 2020.

84. GAVI. Democratic Republic of Congo launches major vaccination drive. [website]. 2018; https://www.gavi.org/news/media-room/democratic-republic-congo-launches-major-vaccination-drive. Accessed 2/27/2020, 2020.

85. World Health Organization. WHO vaccine-preventable diseases: monitoring system. 2019 global summary. 2019. Accessed 7/17/2019.

Chapter 2. Poliovirus Seroprevalence and Vaccine Habits in a Vaccine-Derived Poliovirus Outbreak Site in the Democratic Republic of Congo, 2018

2.1 Abstract

Despite the successes in wild type polio eradication, poor vaccine coverage in the DRC has led to the occurrence of circulating vaccine-derived poliovirus outbreaks. This study provides an update to previous poliovirus seroprevalence studies in the DRC and quantifies risk factors for under-immunization and parental knowledge that guide vaccine decision making. This study is a cross-sectional population-based survey conducted in March, 2018 in eight health zones across Haut Lomami and Tanganyika provinces. Study sites were selected via stratified random sampling in identified health districts using satellite imagery and GIS software. Healthy children between six and 59 months and their parent or guardian were invited to participate in a questionnaire and give a blood sample collection via dried blood spot. Among the 964 participants in our survey, 43.8% (95% CI: 40.6-47.0%), 41.1% (38.0-44.2%), and 38.0% (34.9-41.0%) had protective neutralizing titers to polio type 1, 2, and 3, respectively. We found that 60.7% of parents and guardians reported knowing what polio was, but just 25.6% reported knowing how it spreads. Our data supported the conclusion that polio outreach efforts were successfully connecting with communities—79.4% of participants had someone come to their home with information about polio and 88.5% had heard of a polio vaccination campaign. Additionally, odds of seroreactivity to only serotype 2 was far greater in health zones which had a history of supplementary immunization acitvities (SIAs) compared to health zones that did not: Haut Lomami adjusted odds ratio (aOR) was 6.03 (95% CI: 2.89-12.6); Ankoro & Manono aOR was 5.31 (95% CI: 2.52-11.21). While SIAs may be reaching under-vaccinated communities,

taken together these results are a continuation of the downward trend of seroprevalence rates in this region.

2.1 Background

Progress towards polio eradication is in its final stages. Of the three polio serotypes, currently only wild-type poliovirus (WPV) type 1 is actively circulating in two remining countries: Afghanistan and Pakistan.[1] Globally, the last case of WPV2 was observed in 1999 and in 2015 it was officially declared eradicated. The last case of WPV3 was observed in Nigeria in 2012 and on October 24, 2019, the World Health Organization (WHO) declared it eradicated.[2,3] In the Democratic Republic of the Congo (DRC), importations of WPV1 and WPV3 from India by way of neighboring Angola caused sustained outbreaks from 2006-2011.[4,5] The last confirmed case of WPV3 was confirmed on June 24, 2009 and the last case of WPV1 in Maniema province, DRC was reported on December, 2011.[6-8]

Despite the successes in WPV surveillance and control, poor vaccine coverage in many areas of the DRC has led to the occurrence of circulating vaccine-derived poliovirus (cVDPV) outbreaks.[9,10] VDPVs occur in under-immunized populations when excreted Sabin-strain poliovirus reverts to wild-type virus and experiences community transmission, which can cause paralysis.[11,12] In 2011-2012 there was a large outbreak of 30 cVDPV cases in Haut Lomami province in southeastern DRC.[13] In the wake of the global cessation of the trivalent oral polio vaccine (tOPV) and the resulting immunity gap of polio type 2 among new birth cohorts, areas with low vaccine coverage rates were at increased risk for cVDPV2 outbreaks.[14,15] Indeed, in 2017 cVDPV2 reemerged—a two case cluster was observed in Maniema province and beginning February, 2017, and a 27-case cluster originating in Haut Lomami and spanning three other neighboring provinces was also observed that year.[16] In 2018, 20 cases of cVDPV were reported

from five provinces. Of these, 11 cVDPV2 cases were reported from Mongala province in the northwest of the country. In the southeast, Haut Lomami reported two cases of cVDPV2, Tanganyika reported thee cases of cVDPV2, and Haut Katanga province reported four cases of cVPDV2.[13]

Since 2011 there has been a concentration of cVDPV2 cases in the former Katanga province—now divided into four new provinces as part of DRC's newly-decentralized provincial jurisdictions: Lualaba, Haut-Katanga, Haut Lomami, and Tanganyika provinces—that raise questions about the vaccination landscape of this particular region. Additionally, the dramatic increase in 2018 in case reports from Mongala province in the northwest, speaks to a broader nationwide concern of low poliovirus vaccination rates. On February 13, 2018 the DRC Ministry of Health declared cVDPV2 to be a national public health emergency.[17]

To combat cVDPV emergence, the DRC Ministry of Health in conjunction with the WHO Expanded Programme on Immunization (EPI) coordinates supplementary immunization campaigns (SIAs) in regions where cVDPV2 risk is high or cases have recently been observed. In addition to the vaccines distributed as a part of routine immunization—the bivalent oral polio vaccine (bOPV; introduced April, 2016) and the inactivated polio vaccine (IPV; introduced in 2015)—SIAs employ volunteers in mass door-to-door campaigns which offer additional vaccines to all children under the age of five present in a community, regardless of prior vaccination status.[18] In the DRC, monovalent oral polio vaccine type 2 (mOPV2) or bOPV are used during SIA campaigns.[19]

This study provides an update to previous poliovirus seroprevalence studies that have occurred in the DRC (2014)[7] and in the former Katanga province (2016)[20] and seeks to

understand risk factors for under-immunization and parental knowledge and behaviors that guide vaccine decision making.

2.3 Methods

Study Sample

This study was designed as a cross-sectional population-based survey modeled after the USAID Demographic and Health Survey, with the intention of providing an update on population seroprevalence of markers of poliovirus immunity in an outbreak-prone region of southeastern DRC. Field research was conducted in March, 2018 in eight health zones across Haut Lomami and Tanganyika provinces (HL: Butumba, Lwamba, Malemba-Nkulu, Mukanga; T: Ankoro, Manono, Kabalo, Kongolo). Health zones were chosen and grouped based on the number of cVDPV2 cases and SIAs performed prior to study commencement such that study enrollment was conducted in health zones at different levels risk for cVDPV2 emergence and different recent histories of intervention. Four health zones in Haut Lomami province had a history of cV2DPV cases and had 4-5 SIAs in the past year, Ankoro and Manono health zones in Tanganyika province experienced cVDPV2 cases and had 2 SIAs in the past year, and Kabalo and Kongolo health zones in Tanganyika did not have any cases or SIAs (Figure 2.1).

Within health zones, study sites were selected via a stratified random sampling in identified health districts using satellite imagery-derived settlement feature layers. Villages were randomly selected using ArcGIS software's *Create Random Point* tool using two parameters: selected settlements did not fall in the same administrative Health Area, and had a minimum separating distance of 500 meters. All houses in each selected village were sampled until the necessary sample size was met. Households that refused to participate in the study were marked as refusals

in the tablet-based questionnaire (20/936, 2.14%). This selection method was used to reduce the bias extending from use of microplans, census-like documents used by the EPI, which can unintentionally exclude individual villages or join multiple villages together.

At each eligible household, all healthy children between six and 59 months and their parent or guardian were invited to participate. Study requirements consisted of a questionnaire administered by trained study staff and a blood samples collected via dried blood spot (DBS). Prior to enrollment community leaders were visited at each study site to educate, sensitize, and inform community members about vaccinations and vaccine-preventable disease. Informed consent was administered orally in French, Swahili, or Kiluba by study administrators.

Laboratory Analysis

All collected samples were initially processed at DRC's national reference laboratory in Kinshasa, with one dried blood spot per child shipped to US Centers for Disease Control and Prevention (CDC) for polio testing. The methods used for laboratory analysis of serologic samples have been previously described.[21] Briefly, sera and extracted DBS were processed using the polio microneutralization assay and neutralization titers are reported in a $\log_2$ format, with 2.5 $\log_2$ as the lower limit of detection and 10.5 $\log_2$ as the upper limit of detection. Neutralizing antibodies were assessed against poliovirus serotypes 1, 2, and 3 and titers $\geq$ 3.0 $\log_2$ are considered evidence of seroprotection.

Statistical Analysis

Frequencies, chi-square tests of proportions, and logistic regression models were performed to quantify the relationship between various demographic, knowledge, and behavior

variables and population seroprevalence found to be significant by chi-sq test. While complete polio vaccination requires immunity to all three subtypes, many participants have antibodies to none, some, or all polio serotypes. Owing to the fact that three distinct poliovirus serotypes can cause polio disease and OPV can contain differing combinations of serotype protections, six polio seroprofiles—none, any reactivity to type 1, 2, and 3, seroreactivity to type 2 only, and seroreactivity to all three types—were used as the analytical framework for polio serocoverage. Analyses were performed using SAS version 9.6 (SAS Institute, Cary, NC), maps were generated using ArcGIS software version 10.5 (ESRI, Redlands, CA), and figures were generated using the ggplot2 package (Wickham, 2009) for R (R Core Team, 2014). Ethical approval for this study was obtained via University of California, Los Angeles' Institutional Review Broad (IRB#18-000303) and the Ethics Committee at the Kinshasa School of Public Health in the DRC.

2.4 Results

Among the 964 participants in our survey, 43.8% (95% CI: 40.6-47.0%), 41.1% (38.0-44.2%), and 38.0% (34.9-41.0%) had protective neutralizing titers to polio type 1, 2, and 3, respectively. Seroprevalence varied between individuals: 17.9% (n=172) had neutralizing antibodies for all three polio serotypes, 36.4% (n=351) had none, and 45.7% (n=441) had varying combinations of poliovirus serotypes (Figure 2.2).

In three health zones (Butumba, Malemba-Nkulu, and Mukanga) which were also sampled in a 2016 survey of the region, polio seroprevalence fell an average of 32.6% (range: 16.6-52.0%) (Figure 2.3).

Poliovirus seroprevalence rates increased with age for all serologic profiles except among those who had antibodies to type 2 only. Seroprevalence rates across polio serotypes ranged from 29.8-34.6% for 6-11 month olds, 41.7-48.3% for 12-23 month olds, and 47.4-50.0% for those 24 month or older. Accross each age group type 3 had the lowest seroprevalence (Table 2.1). Notably, number of children in the home and travel time to health facilties, factors often associated with childhood vaccination were not assocaited with polio vaccine seroprevalence.

We also assessed parental knowledge of polio and found that 60.7% of parents and guardians in the study reported knowing what polio was, yet only 25.6% reported knowing how it spreads. When asked about the symptoms of polio, 74.4% of respondents correctly identified paralysis, 38.4% identified fever, and 32.5% identified diarrhea; 14.7% reported that they didn't know the symptoms of poliovirus. Our data found that polio outreach efforts were successfully connecting with communities—79.4% of participants had someone come to their home with information about polio and 88.5% had heard of a polio vaccination campaign (Table 2.2).

Proportions of seroprevalence were similar between the Haut Lomami health zones (n=4), and Ankoro and Manono—the six health zones which experienced cVPDV2 cases and SIAs. These health zones had higher seroprevalence rates for serotype 2 and those with all three antibodies and lower rates of no antibodies compared to Kabalo and Kongolo, health zones which had no cVDPV2 cases or SIAs ($p<.0001$). In Kabalo and Kongolo health zones, 46.1% of the sample population had no poliovirus neutralizing antibodies.

In a logistic regression model estimating seroprevalence of each seroprofile predicted by health zone SIA history and child age, both variables were significant predictors of seroreactivity (Figure 2.4). Increasing age was the positively associated with the odds of seroprevalence with

the exception of those with only markers of type 2 antibodies. The odds of seroreactivity to povliovirus type 2, only type 2, and to all serotypes was increased for healthzones which had cVDPV2 cases and SIA campaigns compared to the two health zones that did not. The odds of seroreactivity to type 2 in Haut Lomami health zones, which had 4-5 SIAs was 4.42 (90% CI: 3.08-6.34) times that of the health zones with none. The odds of seroreactivity to type 2 in Ankoro and Manono health zones, which had 2 SIAs was 4.28 (90% CI: 2.99-6.13) times that of the health zones with none, controlling for other factors. The odds of seroreactivity to only serotype 2 was far greater in health zones which had a history of SIAs compared to health zones that did not: Haut Lomami adjusted odds ratio (aOR) was 5.91 (95% CI: 2.85-12.29); Ankoro & Manono aOR was 5.67 (95% CI: 2.71-11.84).

Seroprevalence of type 1 and type 3 was not associated with the recent SIA history of a health zone. As the number of SIAs increased in a health zone so did the likelihood of both having someone visit your home to distribute information about polio and hearing about a polio campaign. The odds of having a home visit in Ankoro and Manono, health zones that both experienced two SIAs, was 1.06 (95% CI: 0.736-1.53) times that of health zones which had no SIA activity; in Haut Lomami, the odds were 1.90 (95% CI: 1.27-2.84) times greater than the control. Similarly, the odds of hearing about a mass immunization campaign increased with number of campaigns performed in the respondent's residential health zone. In Ankoro and Manono (sites of two SIAs), the odds of hearing of a campaign were 2.25 (95% CI 1.41-3.59) times greater than in control health zones with no SIAs. In the four health zones of Haut Lomami which received four to five SIAs, the odds of having heard about a campaign were 3.59 (95% CI: 2.10-6.13) times greater.

2.5 Discussion

These results are a continuation of the downward trend of seroprevalence rates in this region. In 2014, a national serologic survey observed seroprevalence rates in the former Katanga province (which includes the current Haut Lomami and Tanganyika provinces) of 75-80% for type 1, 85-90% for type 2 and 70-75% for type 3.[7] In 2016, we conducted a serosurvey of the same design in eight health zones in Haut Lomami province (including Butumba, Malemba-Nkulu, and Mukanga surveyed again in this study). That survey found the overall seroprevalence rates to be 79.8% (95% CI: 77.7–81.8%) for type 1, 91.7% (CI: 90.3–93.1%) for type 2, and 70.5% (CI: 68.2–72.8%) for type 3.[20] Since then, overall vaccine coverage rates in this region have fallen to 43.8% (CI: 40.6-47.0%), 41.1% (38.0-44.2%), and 38% (34.9-41.0%) for polio type 1, 2, and 3, respectively.

Similar to our 2016 findings, age was a major predictor of seroprevalence as the older a child is the greater the opportunity for routine vaccination or involvement in an SIA. We also found that only 17.9% of children surveyed had antibodies to all three poliovirus serotypes and many had a diverse mix of seroimmunity profiles, likely a reflection of highly variable and inconsistent polio vaccine distribution in the region.

During routine vaccination with bOPV, type 1 and type 3 are always coupled, thus it was expected that seroprevalence rates of these two serotypes would be roughly equal, however we saw a significantly greater percentage of children seroreactive to type 1 than type 3. This result could be explained by the reduced immunogenicity of serotype 3 in the Sabin vaccine.[22] Additionally, 10.8% of our study (n=104) was seroreactive to only type 2. This was the only serologic profile that was not associated with age, likely indicating that this group was either vaccinated for the first time during a supplementary vaccination campaign via mOPV2 or was

infected with cVDPV2. Overall, the patchwork of vaccination campaigns and use of multiple different OPV vaccines (tOPV, bOPV, and mOPV2) makes tracking and quantifying the overall successes and failures of the cVPDV response in the region difficult. Vaccination cards are often used to track received vaccinations, but in this rural cohort just 13.1% had a vaccination card.

However, regardless of which vaccines have been made available, an increase in the number of SIAs conducted in a health zone was associated with greater overall rates of seroprevalence and also with higher markers of knowledge and outreach activities, such as having someone visit your home to discuss polio or hearing about a vaccination campaign in your area. The association between the number of SIAs conducted in a health zone and seroprevalence rates remained even after controlling for other key factors such as age, sex, and parental knowledge factors. In these rural and semi-rural communities, dissemination of medical resources and information has been an ongoing challenge. One key concern has been that the reports of the number of villages reached and vaccine units distributed may not reflect the fieldwork of a vaccine campaign. A positive relationship between number of campaigns conducted, seroprevalence, and outreach provide useful evidence that SIAs are indeed reaching their target communities and impacting vaccine coverage rates.

While SIAs may be doing their part to improve poliovirus competencies among adults in this region, knowledge of poliovirus and its mechanisms of transmission are still lacking, leaving barriers to community-based prevention and control. Overall, 60.7% of parental guardians in this survey knew of polio disease, but just 25.6% understood how it is transmitted. When asked about the symptoms of polio 74.4% correctly identified paralysis, 38.4% identified fever, and 32.6% identified diarrhea. As paralysis occurs in just 1% of cases[23], there is a pressing need to expand polio understandings to include fever and diarrhea as potential indicators of infection. In the

context of tropical sub-Saharan Africa, this can be a challenge as both symptoms are nonspecific and can indicate infection with a number of other endemic agents, including malaria. However, in a region such as DRC with a history of frequent cVDPV2 cases, community members should be educated to identify persistent fever and diarrhea as potential signs of poliovirus infection.

There were some inconsistencies in parental knowledge of polio. For example, 132 (13.7%) participants identified paralysis as a symptom of polio but responded that they did not know what polio disease was. This could be indicative of different frameworks for disease models that can exist between communities and health practitioners, or a reflection of how polio has been communicated to families by health authorities: as a cause of paralysis rather than an enteric disease. While surveys were administered by local leaders in the local language, perhaps future survey should present these topics in a way more reflective of local conceptions of pathogen and disease.

This study was limited by a few important factors. One key limitation is that the laboratory methods used in this study were not able to distinguish between the presence of neutralizing antibodies due to vaccination or natural infection and the confounding factor that several SIAs were launched in response to cVDPV2 case. Consequently, seroprevalence rates cannot be fully interpreted as a reflection of vaccine coverage. Doing so would likely inflate vaccination rates, particularly for the rates of type 2 since cVDPV2 was circulating in the time leading up to specimen collection. To combat this, we collected vaccine information from vaccine cards which record all vaccinations a child has received.[24] However, as just 13.1% of participants had a vaccine card, preventing the ability to perform a sub-analysis. Other possible limitations include those arising from sampling bias. Yet, as knowledge and outreach factors were also positively associated with number of SIAs conducted in a health zone, we can

conclude that the strength of association between SIAs and seroprevalence is likely not an artifact of reverse causation. We sampled children and their guardians who were present in the village at the time of specimen collection and this strategy may have unintentionally biased our study population. Additionally, we had an underrepresentation of children 35 months or older which, given the strong association between age and seroprevalence, may have lowered our estimates of total seroprotection levels in the study population.

Overall, this survey provides an update to the 2014 and 2016 polio serosurvey conducted in the southeastern Katanga region of the DRC. Since then, polio vaccination coverage rates among children under 5 years of age have fallen to 38-44%. As this region has experienced multiple cVDPV2 outbreaks since 2011 and is a key area for cVDPV2 eradication, a thorough and widespread vaccination strategy is of paramount importance.

2.6 Tables and Figures

Figure 2.1. Map of Haut Lomami & Tanganyika province and study site locations

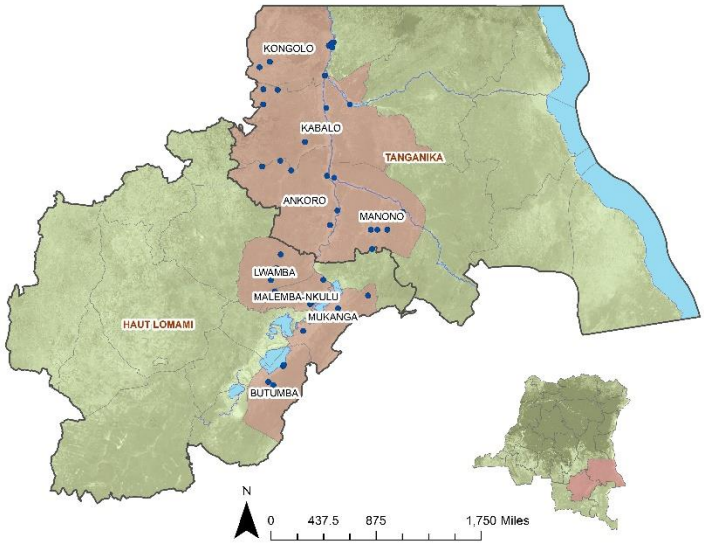


Table 2.1. Participant demographics by poliovirus seroprofile

	Study pop		None		Type 1		Type 2		Type 3		Type 2 Only		All	
	n	Col %	n	Row %	n	Row %	n	Row %	n	Row %	n	Row %	n	Row %
Polio Seroprevalenace														
Type 1	422	43.78	.	.	422	100	260	61.61	279	66.11	.	.	172	40.76
Type 2	396	41.08	.	.	260	65.66	396	100	204	51.52	104	26.26	172	43.43
Type 3	366	37.97	.	.	279	76.23	204	55.74	366	100	.	.	172	46.99
Age														
6 to 11 months	315	32.68	146	46.35	109	34.6	106	33.65	94	29.84	40	12.7	41	13.02
12 to 23 months	609	63.17	191	31.36	294	48.28	268	44.01	254	41.71	62	10.18	118	19.38
24 to 35 months	38	3.94	12	31.58	19	50	22	57.89	18	47.37	2	5.26	13	34.21
Province														
Tanganyika	637	66.08	252	39.56	283	44.43	230	36.11	243	38.15	55	8.63	113	17.74
Haut Lomami	327	33.92	99	30.28	139	42.51	166	50.76	123	37.61	49	14.98	59	18.04
Health Zone SIA History														
4-5 SIAs: Haut Lomami	327	33.92	99	30.28	139	42.51	166	50.76	123	37.61	49	14.98	59	18.04
2 SIAs: Ankoro & Manono	331	34.34	111	33.53	146	44.11	171	51.66	128	38.67	46	13.9	82	24.77
0 SIAs: Kabalo & Kongolo	306	31.74	141	46.08	137	44.77	59	19.28	115	37.58	9	2.94	31	10.13
Sex														
Male	509	52.8	178	34.97	236	46.37	208	40.86	193	37.92	55	10.81	92	18.07
Female	455	47.2	173	38.02	186	40.88	188	41.32	173	38.02	49	10.77	80	17.58
Vaccine Card Present														
Yes	126	13.07	31	24.6	69	54.76	61	48.41	71	56.35	9	7.14	37	29.37
No	554	57.47	169	30.51	278	50.18	247	44.58	225	40.61	58	10.47	106	19.13
Missing	284	29.46	151	53.17	75	26.41	88	30.99	70	24.65	37	13.03	29	10.21
Educational Achievement														
None	190	19.71	70	36.84	68	35.79	83	43.68	69	36.32	33	17.37	31	16.32
Primary school or appretice	551	57.16	201	36.48	237	43.01	216	39.2	207	37.57	59	10.71	92	16.7
Finished secondary school	209	21.68	74	35.41	111	53.11	90	43.06	86	41.15	10	4.78	45	21.53
Higher education	10	1.04	4	40	5	50	6	60	4	40	1	10	4	40
Transportation to Health Facility														
Walk	908	94.19	332	36.56	402	44.27	369	40.64	350	38.55	92	10.13	164	18.06
Boat/Pirogue	22	2.28	9	40.91	8	36.36	12	54.55	4	18.18	5	22.73	4	18.18
Bicycle	21	2.18	4	19.05	7	33.33	11	52.38	8	38.1	6	28.57	2	9.52
Moto	13	1.35	6	46.15	5	38.46	4	30.77	4	30.77	1	7.69	2	15.38
Time to Health Facility														
<10 minutes	376	39	129	34.31	172	45.74	169	44.95	149	39.63	40	10.64	80	21.28
11 to 30 min	308	31.95	120	38.96	125	40.58	119	38.64	119	38.64	32	10.39	52	16.88
30 to 1 hour	135	14	45	33.33	62	45.93	56	41.48	51	37.78	17	12.59	23	17.04
> 1 hour	141	14.63	56	39.72	61	43.26	49	34.75	46	32.62	14	9.93	16	11.35
Dont Know	4	0.41	1	25	2	50	3	75	1	25	1	25	1	25
Number of Children Under 5														
0	10	1.04	3	30	6	60	5	50	6	60	.	.	4	40
1	185	19.19	64	34.59	83	44.86	78	42.16	77	41.62	23	12.43	40	21.62
2-3	598	62.03	213	35.62	260	43.48	249	41.64	229	38.29	67	11.2	106	17.73
4-5	128	13.28	57	44.53	51	39.84	45	35.16	39	30.47	10	7.81	17	13.28
6+	43	4.46	14	32.56	22	51.16	19	44.19	15	34.88	4	9.3	5	11.63

Table 2.2. Guardian knowledge of polio disease (n=964)

	n	%
Do you know what polio is?	585	60.68
Do you know how polio is spread?	247	25.62
If your child was to get sick with polio, what symptoms could they get?		
Paralysis	717	74.38
Fever	370	38.38
Diarrhea	314	32.57
Don't know	141	14.63
What would you do if your child suddenly was unable to walk?		
Take them to a local health care practitioner	452	46.89
Take them to a doctor	245	25.41
Take them to a hospital	133	13.8
I don't know	61	6.33
Do nothing or wait	41	4.25
Treat at home with over the counter medicines	29	3.01
Has someone ever come to your home to give you information about polio?	765	79.36
In the last year have you heard about any Polio Campaigns?	853	88.49
In what ways have received campaign information about polio?		
Community/Village health volunteer	182	18.88
TV commercial	890	92.32
Radio Commercial	948	98.34
Poster	247	25.62

Figure 2.2. Serologic profiles of participants

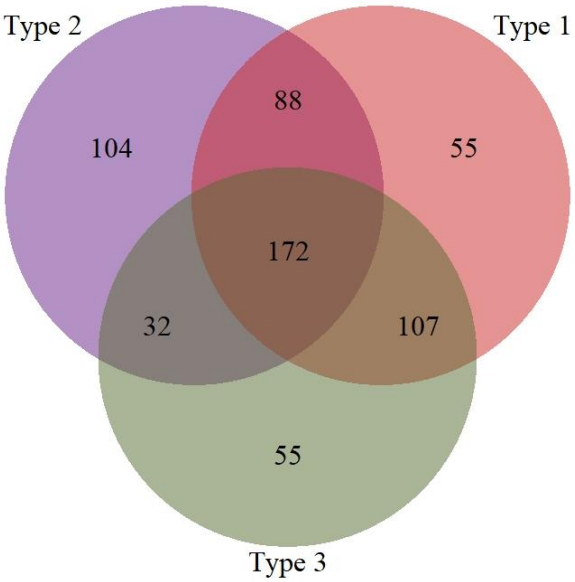


Figure 2.3. Polio seroprevalence, 2016-2018

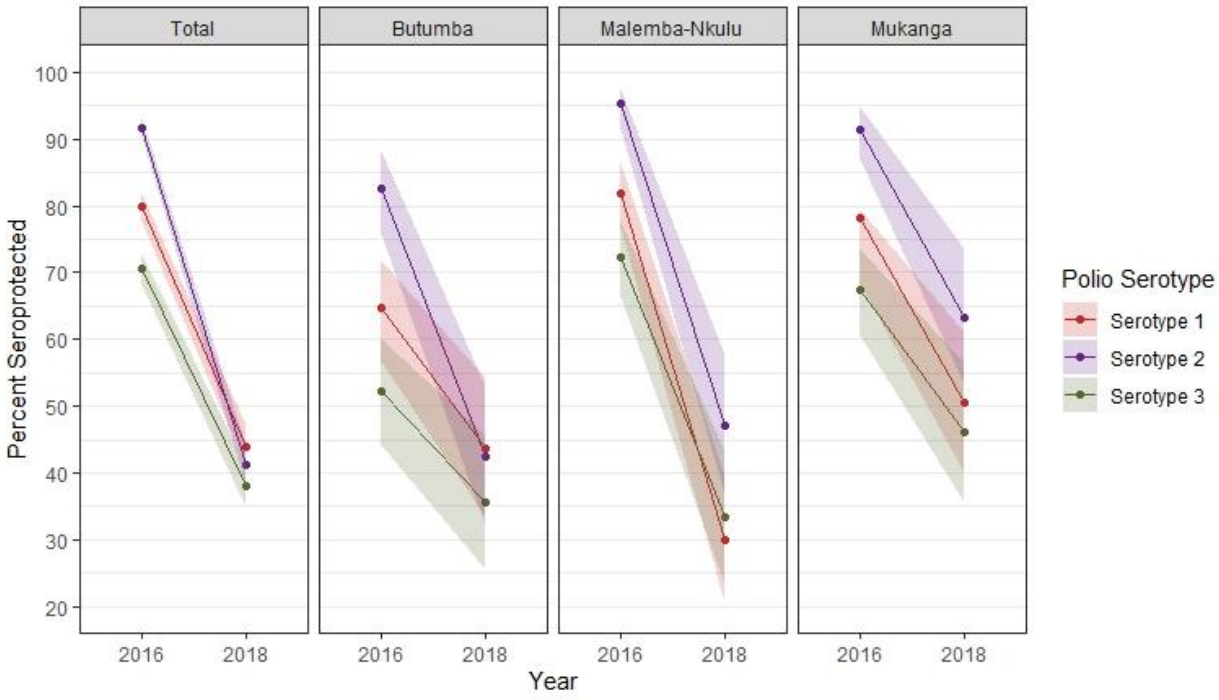
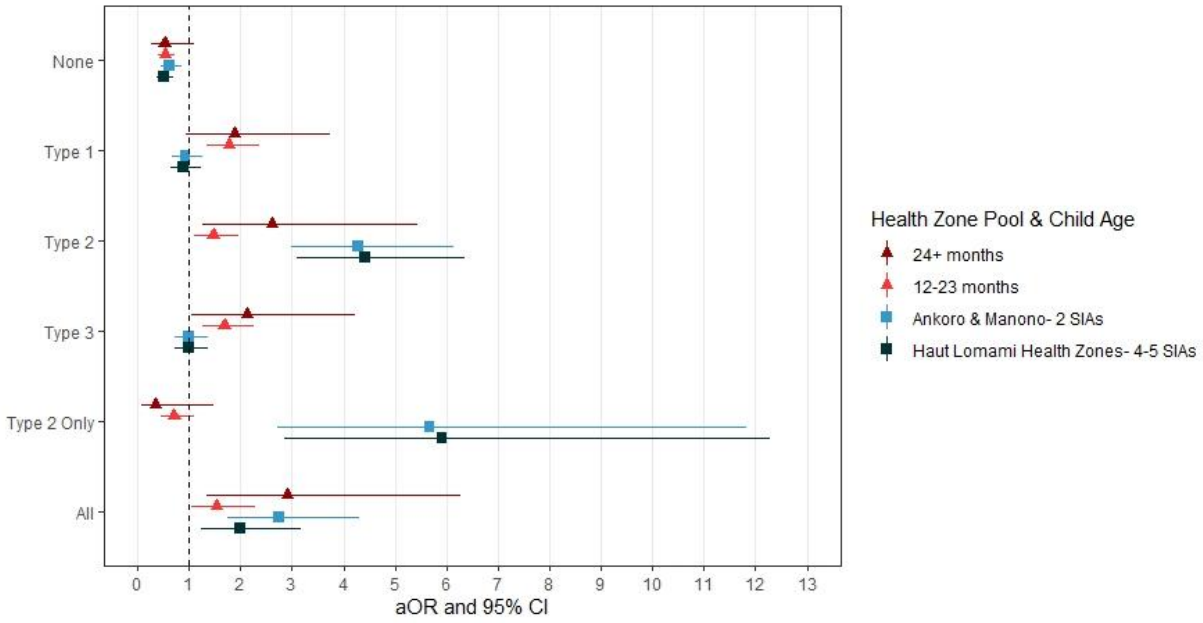


Figure 2.4. Odds of seroreactivity by health zone group



Model used is estimating odds of seroreactivity by health zone SIA history adjusted by age. Reference health zone & age group: Kabalo & Kongolo which had no cVDPV2 cases and 0 SIAs; age 6-12 months.

2.7 References

1. Kabir M, Afzal MS. Epidemiology of polio virus infection in Pakistan and possible risk factors for its transmission. *Asian Pacific Journal of Tropical Medicine.* 2016;9(11):1044-1047.

2. World Health Organization. Two out of three wild poliovirus strains eradicated. 2019; https://www.who.int/news-room/feature-stories/detail/two-out-of-three-wild-poliovirus-strains-eradicated.

3. Kew OM, Cochi SL, Jafari HS, et al. Possible eradication of wild poliovirus type 3—worldwide, 2012. *MMWR Morbidity and mortality weekly report.* 2014;63(45):1031.

4. Centers for Disease Control and Prevention. Wild poliovirus type 1 and type 3 importations--15 countries, Africa, 2008-2009. *MMWR Morb Mortal Wkly Rep.* 2009;58(14):357-362.

5. Gumede N, Jorba J, Deshpande J, et al. Phylogeny of imported and reestablished wild polioviruses in theDemocratic Republic of the Congo from 2006 to 2011. *The Journal of infectious diseases.* 2014;210 Suppl 1(Suppl 1):S361-S367.

6. Burns CC, Diop OM, Sutter RW, Kew OM. Vaccine-Derived Polioviruses. *The Journal of Infectious Diseases.* 2014;210(suppl_1):S283-S293.

7. Voorman A, Hoff NA, Doshi RH, et al. Polio immunity and the impact of mass immunization campaigns in the Democratic Republic of the Congo. *Vaccine.* 2017;35(42):5693-5699.

8. Alleman MM, Chitale R, Burns CC, et al. Vaccine-Derived Poliovirus Outbreaks and Events - Three Provinces, Democratic Republic of the Congo, 2017. *MMWR Morbidity and mortality weekly report.* 2018;67(10):300-305.

9. Wringe A, Fine PEM, Sutter RW, Kew OM. Estimating the Extent of Vaccine-Derived Poliovirus Infection. *PLOS ONE.* 2008;3(10):e3433.

10. Jorba J, Diop O, Iber J, et al. Update on Vaccine-Derived Poliovirus Outbreaks - Worldwide, January 2018-June 2019. *MMWR Morbidity and mortality weekly report.* 2019;68:1024-1028.

11. Fine PEM, Carneiro IAM. Transmissibility and Persistence of Oral Polio Vaccine Viruses: Implications for the Global Poliomyelitis Eradication Initiative. *American Journal of Epidemiology.* 1999;150(10):1001-1021.

12. O'Reilly KM, Lamoureux C, Molodecky NA, Lyons H, Grassly NC, Tallis G. An assessment of the geographical risks of wild and vaccine-derived poliomyelitis outbreaks in Africa and Asia. *BMC Infectious Diseases.* 2017;17:367.

13. Alleman MM, Chitale R, Burns CC, et al. Vaccine-derived poliovirus outbreaks and events—three provinces, Democratic Republic of the Congo, 2017. *Morbidity and Mortality Weekly Report.* 2018;67(10):300.

14. Hampton LM, Farrell M, Ramirez-Gonzalez A, et al. Cessation of trivalent oral poliovirus vaccine and introduction of inactivated poliovirus vaccine—worldwide, 2016. *Morbidity and Mortality Weekly Report.* 2016;65(35):934-938.

15. Membo HK, Mweene A, Sadeuh-Mba SA, et al. Acute flaccid paralysis surveillance indicators in the Democratic Republic of Congo during 2008-2014. *Pan African Medical Journal.* 2016;24(1).

16. Mbaeyi C, Alleman MM, Ehrhardt D, et al. Update on Vaccine-Derived Poliovirus Outbreaks - Democratic Republic of the Congo and Horn of Africa, 2017-2018. *MMWR Morbidity and mortality weekly report.* 2019;68(9):225-230.

17. World Health Organization. Poliomyelitis - Democratic Republic of the Congo. *Disease outbreak news* 2018; https://www.who.int/csr/don/20-December-2018-poliovirus-drc/en/. Accessed 2/27/2020, 2020.

18. Helleringer S, Abdelwahab J, Vandenent M. Polio supplementary immunization activities and equity in access to vaccination: evidence from the demographic and health surveys. *The Journal of infectious diseases.* 2014;210(suppl_1):S531-S539.

19. Global Polio Eradication Initiative. mOPV2 Management, Monitoring, Removal and Disposal. In:2021.

20. Halbrook M, Alfonso VH, Hoff NA, et al. Vaccination and serologic response against poliovirus among children in an outbreak area of vaccine derived poliovirus in the Democratic Republic of the Congo. In:2020.

21. Weldon WC, Oberste MS, Pallansch MA. Standardized Methods for Detection of Poliovirus Antibodies. *Methods in molecular biology (Clifton, NJ).* 2016;1387:145-176.

22. Reinheimer C, Friedrichs I, Rabenau HF, Doerr HW. Deficiency of immunity to poliovirus type 3: a lurking danger? *BMC Infectious Diseases.* 2012;12(1):24.

23. Nathanson N, Kew OM. From Emergence to Eradication: The Epidemiology of Poliomyelitis Deconstructed. *American Journal of Epidemiology.* 2010;172(11):1213-1229.

24. Miles M, Ryman TK, Dietz V, Zell E, Luman ET. Validity of vaccination cards and parental recall to estimate vaccination coverage: a systematic review of the literature. *Vaccine.* 2013;31(12):1560-1568.

Chapter 3. Impact of Supplementary Immunization Campaigns on Child Polio Vaccination Status in Eastern DRC, 2016-2019

3.1 Abstract

Supplementary immunization activities, like door-to-door vaccine campaigns, are a key public health intervention in areas where polio vaccine coverage is low and vaccine-derived polio cases persist. Conducting these campaigns in low-resource and remote areas presents many challenges to vaccination teams. Quantifying the impact of a vaccine campaign on population seroimmunity provides key information for polio eradication strategy. This study utilized data collected from three cross-sectional representative panel surveys conducted from 2016-2019 across seven health zones in Haut Lomami province, DRC. Fixed effects regression models and inverse probability treatment weighting were used to quantify the relationship between participation in a vaccine campaign and a child's seroimmunity and vaccination status. The sensitivity of parental recall of child immunization to identify serology, a gold standard method for detecting vaccination status was calculated. This study includes biological specimens and questionnaire data from 1,227 children and their guardians collected in 2016, 152 children in 2018, and 430 children in 2019. Guardian recall of childhood vaccination status had a sensitivity of 76.8% and specificity of 50.5% when compared to the serologic evidence of vaccination. The odds of seropositivity to polio type 2 increased by 1.07 (95% CI: 1.00-1.15) for each SIA a child had ever participated in, controlling for age and having a health worker visit your home. For each increase in the number of SIAs a child participated in, the odds of being fully vaccinated increased by 1.42 (95% CI: 1.34-1.50), controlling for age and having someone visit your home to discuss polio. This study reports from some of the first longitudinal seroprevalence studies in this high-risk region for cVDPV2 emergence. While it appears that over time opportunity to

participate in an SIA increased, declining reports of participation coupled with declining rates of seroprevalence and vaccine coverage and an increase in cVDPV2 cases suggests that SIAs may struggle to reach portions of the population.

3.2 Introduction

Poliovirus eradication faces a unique challenge due to the nature of the oral polio vaccine (OPV). OPV, or the Sabin vaccine, uses attenuated poliovirus to confer robust humoral and mucosal immunity, protecting an individual from paralysis and interrupting transmission.[1,2] However, in rare instances and in environments where vaccine coverage is low, Sabin polio strains can undergo genetic changes and result in circulating vaccine-derived poliovirus (cVDPV), which can cause paralytic polio.[1] Since 2000, when OPV was discovered as the source of a polio outbreak, cVDPV risk has been a major obstacle to achieving eradication through vaccination.[3-8] Ultimately, to achieve poliovirus eradication, full OPV cessation will be required.[9] In many industrialized nations, this has already occurred by replacing OPV with the inactivated polio vaccine (IPV), or Salk vaccine, which confers humoral immunity, but not mucosal immunity and thus cannot interrupt transmission.[10,11] While wild poliovirus type 2 (WPV2) has not been observed since 1999, cases of cVDPV2 persist due to the use of the trivalent OPV (tOPV), which contains polio types 1, 2, and 3. As part of the long-term eradication strategy, in April 2016, the Global Polio Eradication Initiative (GPEI), an initiative supported by the WHO, UNICEF, and The Gates Foundation coordinated a global effort to switch from tOPV to a bivalent OPV (bOPV) containing types 1 and 3 only.[12,13] While some cVDPV2 cases were expected following the global switch, VDPV2 genomic surveillance suggests that over half of the detected isolates of cVDPV2 had a high probability of being seeded after the switch.[5]

Current polio endgame strategy in areas where cVDPV cases are detected relies on supplemental immunization activities (SIAs) to improve vaccine coverage to mitigate cVDPV risk.[14] These SIAs are conducted in addition to childhood routine immunization and target geographic areas of high risk during either national immunization days (NID), subnational immunization days (SNID), mop-up campaigns, or case response campaigns.[15] To combat cVDPV2 infection and subsequent AFP cases, monovalent OPV type 2 (mOPV2) is often used during an SIA. However, the epidemiology of Sabin strains and VDPV has created a paradoxical situation for polio endgame strategy. Vaccination remains the greatest tool for polio control, yet insufficient coverage and overuse of mOPV2 drives cVDPV2 emergence. Indeed, improving the quality of SIAs for VDPV prevention was listed as a key challenge in the WHO's updated 2019-2023 Polio Endgame Strategy.[14]

The Democratic Republic of the Congo (DRC) has documented cases of VDPVs since 2004 with outbreaks recorded in 2011-2012 and from 2017 to present day.[16-19] The overall paradigm for SIAs are designed and regulated by GPEI and have been a critical polio eradication strategy in DRC since 1996.[20] In a low-resource and logistically challenging nation like the DRC, SIAs, organized and administered by the DRC's Expanded Programme on Immunization (EPI) and Ministry of Health, face real challenges to reach a high proportion of the population.[21] Two key facets of SIA planning are the microplan and cold-chain logistics. For SIA microplanning, the nation's 26 provinces are divided into smaller administrative areas called health zones. All villages within a health zone are enumerated and population counts are gathered from local leadership.[22] Currently, the DRC is experiencing a time of transition from hand-drawn maps to digital satellite based-imagery and a piecemeal population update to the previous national census last run in 1984. Achievements in population record keeping and

mapping are improving SIA planning efforts, but complete estimations of population size and village locations are essential for successful vaccine intervention. Additionally, SIA planning must organize the logistics of deploying vaccines from the centralized storage facility to individual health facilities while maintaining proper cold chain. OPV must be kept between 2° and 8° Celsius during transit. Once deployed, vaccinators use a combination of house-to-house and fixed-post strategy for vaccine delivery but in many parts of the country where electricity is inconsistent, neither strategy can ensure proper cold storage of OPV.[13] One study among vaccination teams conducted during a NID in Mali found that 53.1% of children vaccinated in the campaign received OPV that was kept outside of the cold chain yet over the course of each day vaccine vial monitors did not detect vaccine spoilage. Vaccinators interviewed in this study reported preferring working outside of the cold chain due to the difficulties incurred by carrying ice packs.[23] However, other studies have questioned the reliability of vaccine vial monitors and caution placing complete reliance solely on them.[24]

Despite careful SIA planning and teams of dedicated health workers, community seroprevalence in DRC has declined. GEPI calendars of SIA events in the DRC record many SIAs targeting high risk areas, yet polio serocoverage remains below target levels and cVDPV2 cases persist. In VDPV-endemic regions, true understanding of population immunity against polioviruses requires population serosurveillance, but such studies can be costly and resource-intensive.[25] Additionally, while often accurate, the use of serologic data as evidence for polio vaccination is susceptible to reverse causation, particularly in outbreak-prone regions as standard serologic methods are unable to differentiate between infection and vaccination. Community based household studies, like the Demographic and Health Survey (DHS), assess vaccination history based on physical records, like vaccine cards, or parental recall. These data likewise are

susceptible to bias including selection bias, information bias, data entry errors, and missing data.[26,27] Accurate estimates of vaccine coverage are paramount, and understanding the associations between vaccine recall and serologic biomarkers as measures of immunity can help improve epidemiologic surveillance of community vaccination in low resource environments.

We conducted fieldwork in seven heath zones in Haut Lomami province between 2016 and 2019 in order to quantify polio antibody seroprevalence rates, childhood vaccine coverage, and to further measure the community impact of SIAs on polio immunity over time.

3.3 Methods

Poliovirus seroprevalence studies have been performed in southeastern DRC in 2016[28], 2018[29], and 2019. During these surveys, the Kabando-Dianda *antenne*, a group of health zones representing a vaccine distribution region, has been visited twice. These seven health zones—Bukama, Butumba, Kabondo-Dianda, Kinkondja, Malemba-Nkulu, Mukanga, and Mulongo—were first visited in 2016 and then revisited in either 2018 or 2019 (Figure 3.1).

Sampling Methods

These seroprevalence studies are cross-sectional representative panel surveys. The sampling methodology for each serosurvey has been described in detail elsewhere[28,29] but briefly, each survey employed a two-stage sampling design for participant selection. In the first stage, enumeration areas (EAs)—neighborhoods in larger cities, whole of smaller cities, and sectors or *chefferies* in rural districts— were listed from either available records or satellite imagery and chosen randomly such that chosen EAs didn't fall in the same health area (an administrative unit at the sub-health zone level) and had a minimum separating distance of 500 meters. In the second phase, all households with a child under either 48-months (2016 survey) or

24-months (2018 & 2019 surveys) and consenting guardian present at the time were invited to participate. Prior to enrollment, community leaders visited each study site to educate, sensitize, and inform community members about vaccinations and vaccine-preventable disease. During enrollment consent was administered orally in the local dialect (French, Swahili, or Kiluba) by study administrators.

Data Collection

Interviewers were selected from the community and trained to administer the questionnaire using Open Data Kit 2016 version 2.0.4 (UW-CSE, Seattle, WA) on tablets. If the mother was available, preference was given to her and if not, another familial guardian was interviewed. Data was collected on household information, guardian demographics, knowledge and attitudes related to polio, and each child's vaccination history. If available, each child's vaccination card was recorded. Dried blood spot samples were collected via finger or heel prick for children under 12 months using disposable safety lancets and protein saver cards.

Parents were asked about a child's vaccination status to the four recommended OPV vaccines and the IPV vaccine given at birth, 6 weeks, 10 weeks, and 14 weeks and the number of times their child participated in an SIA campaign. In addition to parental recall to assess lifetime SIA vaccination, a variable summarizing the number of SIA campaigns run in each health zone in the year prior to the 2016 survey and in the months between the 2016 serosurvey and the 2018 or 2019 surveys was calculated (SIA schedule included in Appendix 3.1). Information for this variable was provided via surveillance data[30] and situation reports made available via the DRC EPI.

Laboratory Analysis

The US Centers for Disease Control and Prevention (CDC) tested all useable samples for neutralizing antibodies against poliovirus serotypes 1, 2, and 3.[31] Neutralization titers are reported in a log2 format, with 2.5 log2 as the lower limit of detection and 10.5 log2 as the upper limit of detection. Neutralizing antibodies were assessed against poliovirus serotypes 1, 2, and 3 and titers ≥ 3.0 log2 are considered as evidence of seroprotection. Analysis of blood specimens were available for data collected in 2016 and 2018.

Data Analysis

In accordance with CDC guidelines, children were classified as fully vaccinated against poliovirus if their guardian confirmed vaccination with at least three doses of vaccine[32]; children with just one or two doses of vaccine were classified as partially vaccinated. Data was compiled and analyzed using SAS 9.4 (Cary, NC) and figures were created in R (R Core Team, 2014) using the ggplot2 package (Wickham, 2009).

Chi-square test of proportions and trend tests were used to analyze associations between key variables, polio vaccination status, lifetime SIA participation, a health zone's recent SIA history, and year of survey. To estimate the effect participation in an SIA had on vaccination status, a fixed effects logistic regression model was employed. As guardian recall of childhood vaccination in this population can be inaccurate, blood samples measuring polio seroreactivity can be considered to reflect a child's true vaccine status. Sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) were calculated to estimate the how measuring vaccination by guardian recall compares against using a serologic biomarker. A sensitivity model (Model 1) comprised of data from 2016 and 2018 measures the impact of SIAs on polio seroreactivity. The relationship between SIA participation and reported vaccination status was explored in Model 2 using data from all three years. To adjust for outcome

misclassification arising due to imprecise recall of vaccination status a propensity score was calculated to estimate the probability of being seropositive based on relevant cofounders and used vaccine recall model as an inverse probability treatment weight (IPTW, Model 2).

Propensity score estimation:

$$e_i = P(Y_i = 1 \mid age_i, health\ zone_i, VDPV\ cases_i, SIA\ doses_i,)$$

Finally, in models 1 and 2, participation in an SIA was measured two different ways. It was included as an ecological predictor—as the number of SIAs run in the health in the period prior to sample collection (Exposure a) or as an individual level predictor (Exposure b) measuring the number is SIAs a child had reportedly participated in.

3.4 Results

Sample size

This study includes biological specimens and questionnaire data from 1,227 children and their guardians collected in 2016, 152 children in 2018, and 430 children in 2019. Across the seven health zones included, the average enrollment was 258 children per year (range: 184-331). In total, 67.9% of children in this study reported being fully vaccinated against poliovirus; 12.4% had partial vaccination and 19.7% had no reported vaccinations. Across study years, the proportion of participants reporting full vaccination decreased (77.6%, 52.6%, 45.6% in 2016, 2018, 2019, respectively) while the proportion of children with no vaccination increased (13.2%, 24.3%, 36.7%, in 2016, 2018, 2019, respectively). Accordingly, rates of recorded seroprevalence dropped between 2016 and 2018 (Figure 3.2). Guardian recall of childhood vaccination status had a sensitivity of 76.8% and specificity of 50.5% when compared against the serologic

evidence of childhood vaccination. Additionally, the positive predictive value of guardian recall was 88.5% and a negative predictive value of 30.5%.

Reported participation in an SIA campaign decreased over time (Kruskal-Wallis p-value <.0001). In 2016, guardians reported that their child had participated in a lifetime average of 9.53 (95% CI: 9.27-9.80) SIAs. In 2018, the average dropped to 5.36 (95% CI: 4.88-5.83) and in 2019 the average number SIAs that children participated in in their lifetimes was 3.58 (95% CI: 3.37-3.80). Across the seven health zones, official records indicated that there was a range of 5-12 SIAs performed in a health zone in the periods prior to investigation. When asked how many SIAs a child had ever participated in, guardians responded with a wide range of values between 0-19.

In each survey period neither sex nor age was associated with vaccination status. Rates of vaccination differed across health zones in 2016 and in 2018/2019. In both time periods Kabando-Dianda had the highest rate of fully vaccinated children (17.75% and 34.18%, respectively). The number of SIAs a child has ever participated in was associated with vaccination completeness in each year. In 2016, 60.6% of children who reported full vaccination also reported participating in nine or more SIA campaigns. In 2018/2019, 57.3% of those fully vaccinated, reported participating in 4-8 SIAs. Many children reported never receiving a polio vaccination, but also had participated in several SIAs. In 2016, 65.4% (n=106) of children reporting no vaccination had also participated in four or more SIAs. As SIA participation increased, so did the proportion of participants reporting full vaccination. Variables associated with vaccine outreach such as hearing of a polio vaccination campaigns in the past year were positively associated with vaccination status. A majority of participants had heard of vaccination campaigns run in their area regardless of the child's vaccine status—in 2016 93.4% of

participants had—but in 2018/2019 this proportion dropped to 82.6%. Among the 17.4% of participants who hadn't heard of a campaign, 73.0% (n=73/100) reported their children did not have any vaccinations against poliovirus. Additionally, in 2018/2019 having a health worker visit your home, was strongly associated with vaccine status (Table 3.1).

In Model 1, which estimates the association between seropositivity to serotype 2 and SIA participation between 2016 and 2018, found that compared to the five SIAs each health zone received prior to data collection at timepoint 1, the 2-4 additional SIAs ran between timepoint 1 and 2 had a negative impact on type 2 seroprevalence, controlling for age and having a health worker visit your home. The odds of seropositivity to type 2 among health zones which received 7-8 SIAs was 0.13 (95% CI: 0.06-0.27) times that compared to those which received five. However, when classifying SIAs as lifetime participation, the odds of seropositivity to type 2 increased by 1.07 (95% CI: 1.00-1.15) for each SIAs a child participated in, controlling for age and having a health worker visit your home (Table 3.2).

In Model 2a, which estimates the association between full vaccination and SIAs, the previous SIA record for a health zone was negatively associated with vaccination completeness and did not display a monotonic trend as number of SIAs increased. The number of SIAs a child had ever participated in (Model 2b) was positively associated with vaccination status (Figure 3.3). For each increase in the number of SIAs a child participated in, the odds of being fully vaccinated increased by 1.42 (95% CI: 1.34-1.50), controlling for age and having someone visit your home to discuss polio.

3.5 Discussion

This study reports from some of the first longitudinal seroprevalence studies in this high-risk region for cVDPV2 emergence. Across the study period both vaccination and

seroprevalence decreased among children under five years of age. As vaccination coverage has decreased, cVDPV2 cases in the Haut Lomami province and DRC in general have increased from 26 cases nationwide, 8 cases in Haut Lomami in 2016 to 89 cases of cVDPV2, 19 in Haut Lomami in 2019. Notably, 2020 reports recorded 74 cVDPV2 cases nationwide, none of which were in Haut Lomami province. While DRC Ministry of Health situation reports recorded that all health zones in the Haut Lomami had reported their VDPV cases, the COVID-19 pandemic may have interfered with surveillance and reporting systems.

While cVDPV2 cases increased and seroprevalence decreased, participants reported a decreasing involvement in SIAs over time. Records on when and where SIAs were conducted in these health zones from this time indicate that there were three sub-national immunization days in 2015, two national immunization days in 2016, and then an expansion in 2017 to four SIAs which covered all health zones and an additional three SIAs which targeted particular health zones of high risk. In 2018, records indicate that six SIAs were run in all seven health zones of study interest. While it appears that over time opportunity to participate in an SIA increased, declining reports of participation coupled with declining rates of seroprevalence and vaccine coverage and an increase in cVDPV2 cases suggests that SIAs may struggle to reach portions of the population. We observed the highest rates of reported full vaccination in Kabando-Dianda health zone. As Kabando-Dianda is the resource hub for this antenne, higher rates in this region might reflect the ways that vaccine access is impacted by national vaccine supply chain logistics—vaccine arrives first in Kabando-Dianda before heading out to surrounding health zones.

In the serologic model, we observed that the SIAs ran in Butumba, Malemba-Nkulu, and Mukanga health zones did not improve seroprevalence of serotype 2. Of the nine SIAs ran

between our survey timepoints in these health zones, five used vaccine containing serotype 2. Interpretation of this finding is further complicated, however, by the collinearity between the number of SIAs conducted and time as well as the potential for reverse causation as each of these health zones experienced cVDPV2 cases, which impacts both SIA scheduling and individual seroimmunity. Increased participation in an SIA across a child's lifetime, while susceptible to recall bias, was associated increased seroprevalence to type 2. The recall model reported similar trends as the serologic model—the SIA record of a health zone was negatively associated with full vaccination status, but lifetime SIAs was positively associated with full vaccination.

CDC defines full vaccination as having three or more doses of poliovirus vaccination. According to governmental record, all seven health zones in this investigation received more than three SIAs and parental recall reported that 77.2% of children had participated in three or more SIAs over their lifetime. Yet, seroprevalence rates in the region are below target levels and just 13.7% of guardians had vaccine cards. These data problematize the relationship between supplemental immunization activities and routine immunization efforts for children under five and how, while SIA coverage reports are high, population seroimmunity rates trail behind. While lifetime SIA participation was associated with full vaccination, the strength of association suggests that SIAs could improve their community impact. Aside from national immunization days, which are a part of the global effort to improve vaccination, mop-up and case response campaigns respond directly to cVDPV2 outbreaks and focus exclusively on improving serocoverage for polio serotype 2. We found that seroprevalence to serotypes 1 and 3 lag behind serotype 2, likely due to this frequent use of mOPV2.[28,29] While tOPV hasn't been in use since April 2016, increased exposure to vaccine containing serotypes 1 and 3 may help improve overall population coverage rates.

Additionally, this study sought to compare two methods for measuring vaccination coverage. While the serologic record is often thought as a gold standard, questionnaire-based data, like guardian recall of a child's vaccination status is often more feasible to collect. A sensitivity analysis estimated that among this cohort where confirmatory vaccine records are scant, parental recall was able to correctly identify 76.9% of seropositive participants, and 50.5% of persons who were seronegative. This discordance could be interpreted as an outcome misclassification. While much epidemiologic research is focused on misclassification or mismeasurement of an exposure far less attention has been given to misclassification of an outcome.[33-35] Here the use of a propensity score was adopted to reduce the bias due to outcome mismeasurement.[36] Both propensity score regression adjustment and IPTW methods were tested, and while using an IPTW performed better, neither method was able to fully eliminate the bias due to outcome misclassification.[37,38] This may be partially explained by an incorrect specification of the propensity score model, further research is necessary to quantify the bias incurred while measuring outcomes other than identified gold standard biomarkers.

Importantly, current laboratory methods are unable to distinguish between the presence of antibodies due to VDPV infection or vaccination. As this is a cVDPV2 outbreak region, using the serologic record as evidence for vaccine coverage may lead to overestimations of how vaccine programs are performing. Measuring the effectiveness of SIAs are further complicated by temporal issues as they are both activated in response to a VDPV case and as prevention against future cases. Given this, the question of how SIAs perform in the context of a VDPV-endemic region faces many collinearity issues in the data most likely to be collected, as was the challenge here. Future work on this question should seek out study designs which can adequately address these issues. For example, in this study just seven health zones were visited twice. A

larger longitudinal panel study, which visited more health zones, or an individual-level repeated measured design could have sufficient k-level groups to employ a multilevel logistic regression model, thus able effectively separate the impact of SIA from location and timepoint.

Commitment to high quality vaccine coverage in this rural, logistically challenging region remains a public health challenge. Multiple data sources report that children under five are interfacing with multiple supplemental vaccine campaigns, yet seroprevalence and vaccine coverage remain subpar. While the frequency of vaccination in this region should remain, efforts should be made to improve vaccine access to those living in hard-to-reach areas, improve record keeping, and increase the impact a single immunization campaign has on population seroprevalence.

3.6 Tables and Figures

Figure 3.1. Study Region by Year

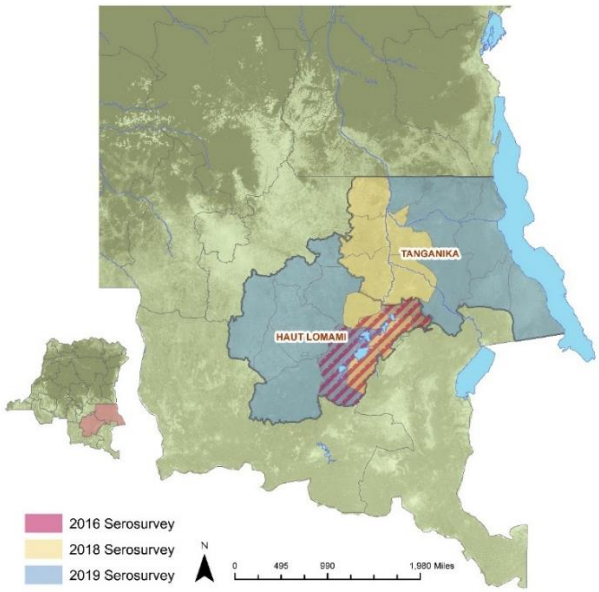


Figure 3.2. Vaccination Recall and Seroprevalence, by Year

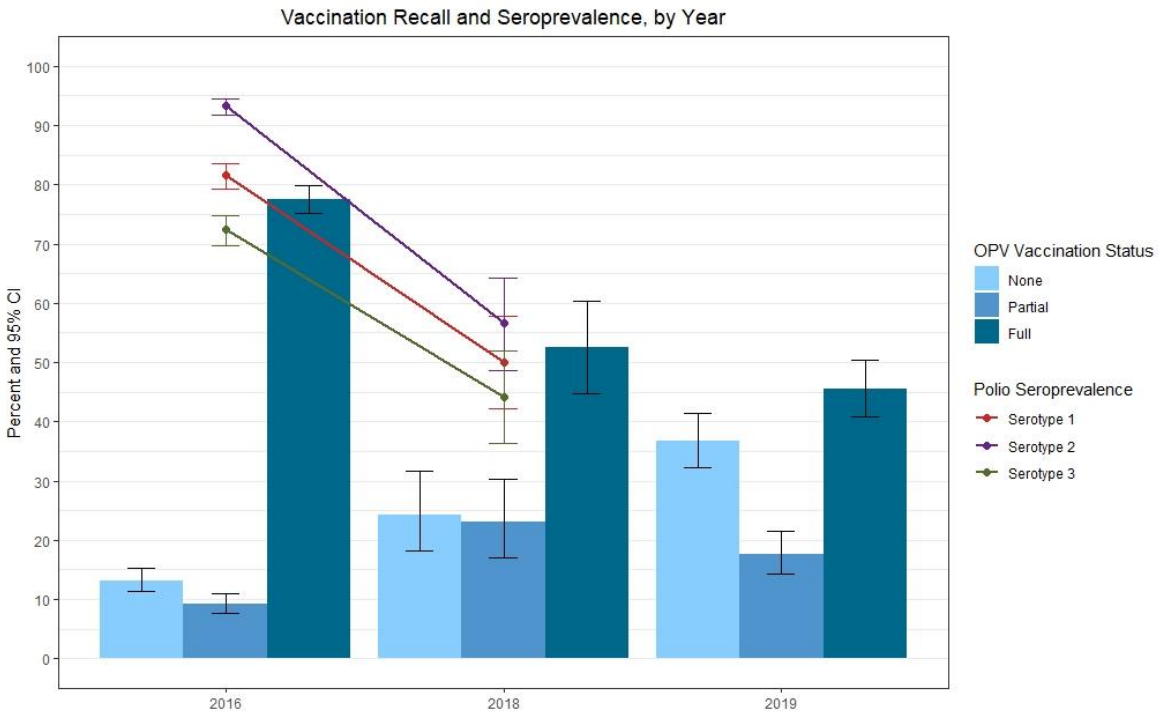


Table 3.1. Participant demographics, by year and vaccination status

	Time 1: 2016 (N=1227)						Time 2: 2018-2019 (N=582)					
	No Vaccinations n=162		Partial Vaccination n=113		Full Vaccination n=952		No Vaccinations n=195		Partial Vaccination n=111		Full Vaccination n=276	
	μ	σ	μ	σ	μ	σ	μ	σ	μ	σ	μ	σ
Age (months)*	36.7	15	33.8	15.9	35.9	16.1	15	5.8	13.9	5.4	13.7	5.4
	n	Col %	n	Col %	n	Col %	n	Col %	n	Col %	n	Col %
Sex												
Male	86	53.09	64	56.64	503	52.84	102	52.31	62	55.86	168	60.87
Female	76	46.91	49	43.36	449	47.16	93	47.69	49	44.14	108	39.13
Health Zone												
Bukama	32	19.75	23	20.35	113	11.87	36	18.46	19	17.12	55	19.93
Butumba	28	17.28	20	17.7	84	8.82	13	6.67	9	8.11	30	10.87
Kabondo-Dianda	7	4.32	11	9.73	169	17.75	28	14.36	11	9.91	67	24.28
Kinkondja	33	20.37	9	7.96	122	12.82	43	22.05	7	6.31	55	19.93
Malemba-Nkulu	13	8.02	15	13.27	160	16.81	19	9.74	17	15.32	28	10.14
Mukanga	15	9.26	13	11.5	138	14.5	5	2.56	9	8.11	22	7.97
Mulongo	34	20.99	22	19.47	166	17.44	51	26.15	39	35.14	19	6.88
Vaccine card present												
Yes	5	3.09	23	20.35	103	10.82	2	1.03	29	26.13	86	31.16
Lifetime SIA Participation												
0-1	19	11.73	16	14.16	46	4.83	101	51.79	40	36.04	25	9.06
2-3	37	22.84	24	21.24	59	6.2	52	26.67	44	39.64	75	27.17
4-8	71	43.83	54	47.79	270	28.36	40	20.51	26	23.42	158	57.25
9+	35	21.6	19	16.81	577	60.61	2	1.03	1	0.9	18	6.52
SIA Record*												
5	162	100	113	100	952	100						
7							13	6.67	9	8.11	30	10.87
8							19	9.74	17	15.32	28	10.14
9							5	2.56	9	8.11	22	7.97
11							122	62.56	57	51.35	141	51.09
12							36	18.46	19	17.12	55	19.93
Heard of polio campaign in past year												
Yes	145	89.51	102	90.27	899	94.43	121	62.05	103	92.79	257	93.12
Polio home visit in last year												
Yes	133	82.1	99	87.61	846	88.87	114	58.46	97	87.39	248	89.86

*surveys from time 1 included ages from 6-59 months; surveys from time 2 included ages 6-23 months

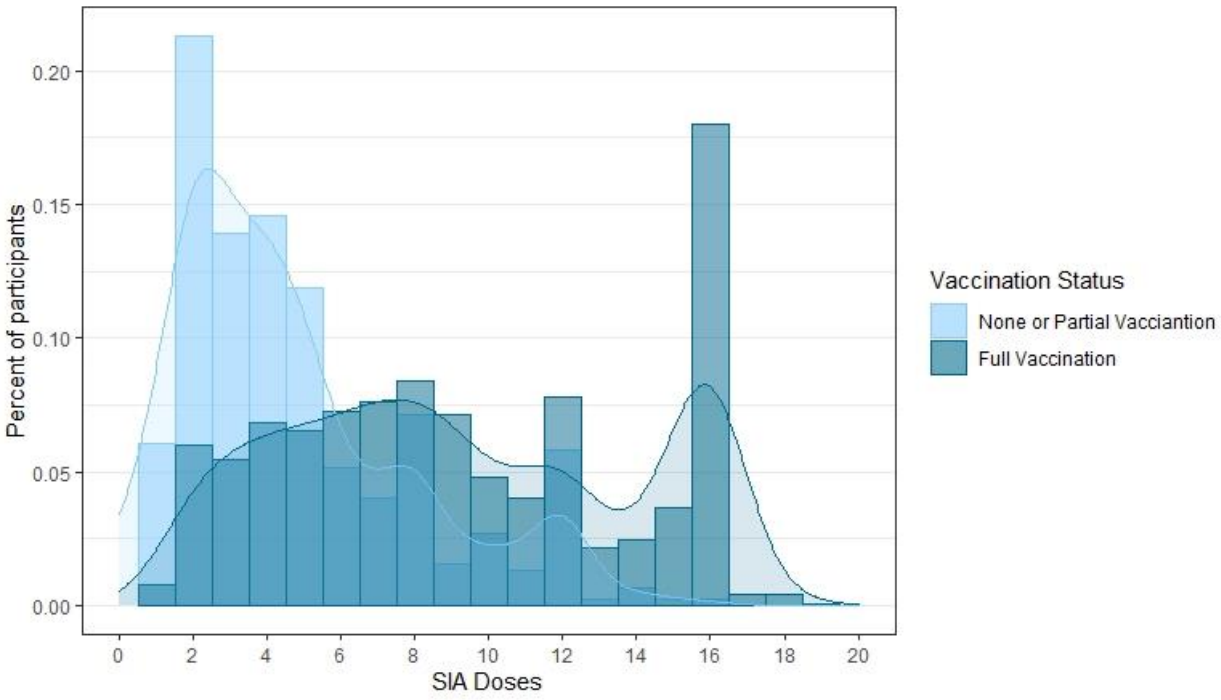
Table 3.2. Modelling association between SIA exposures and measures of childhood vaccination

SIA Measure	Model 1- Serology Model					Model 2- Recall model*				
	Variable		**aOR**	**95% CI**		**Variable**		**aOR**	**95% CI**	
a) Number SIAs run in period prior to study collection**	SIA Record	5	*ref*	*ref*	*ref*	SIA Record	5	*ref*	*ref*	*ref*
		7	0.13	0.06	0.27		7	0.43	0.22	0.87
		8	0.13	0.07	0.26		8	0.24	0.13	0.43
		9	0.73	0.26	2.02		9	0.63	0.26	1.51
	Age (months)	cont.	1.03	1.00	1.05		11	0.50	0.31	0.81
	Polio home visit in last year	No	*ref*	*ref*	*ref*		12	0.41	0.24	0.71
		Yes	1.25	0.55	2.84	Age	cont.	1.02	1.01	1.03
						Polio home visit in last year	No	*ref*	*ref*	*ref*
							Yes	0.81	0.37	1.73
b) Lifetime child SIA doses	SIAs ever	cont.	1.07	1.00	1.15	SIAs ever	cont.	1.42	1.34	1.50
	Age (months)	cont.	1.04	1.02	1.06	Age (months)	cont.	0.97	0.95	0.98
	Polio home visit in last year	No	*ref*	*ref*	*ref*	Polio home visit in last year	No	*ref*	*ref*	*ref*
		Yes	1.11	0.50	2.46		Yes	0.52	0.20	1.40

* weighted with propensity score to adjust for bias

** see Appendix Table 1 for full schedule of SIAs and study collection dates

Figure 3.3. Histogram of SIA Doses by Vaccination Status



3.7 Appendix

Appendix Table 3.1. SIA schedule and study collection dates

	2015			2016			2017										2018					2019	
	SNID tOPV Apr 30	SNID tOPV Nov 5	SNID tOPV Dec 3	NID tOPV Mar 24	NID tOPV Apr 14		NID bOPV Apr 9	CR mOPV2 Jun 6	SNID mOPV2 Jul 13	Mop up mOPV2 Sep 14	SNID bOPV Oct 12	SNID mOPV2 Nov 30	SNID mOPV2 Dec 16	SNID bOPV Jan 6	NID bOPV Jan 8		SNID mOPV2 Apr 26	Mop up mOPV2 Jun 26	CR mOPV2 Sep 27	CR mOPV2 Oct 11	CR mOPV2 Jan 31	NID bOPV Apr19	
BUKAMA	x	x	x	x	x		x		x			x	x	x	x		x	x	x	x	x	x	
BUTUMBA*	x	x	x	x	x	*Survey July*	x	x	x			x	x	x	x	*Survey March*	x	x	x	x	x	x	*Survey June*
KABONDO-DIANDA	x	x	x	x	x		x		x			x	x	x	x		x	x	x	x		x	
KINKONDJA	x	x	x	x	x		x		x			x	x	x	x		x	x	x	x		x	
MALEMBA-NKULU*	x	x	x	x	x		x	x	x		x	x	x	x	x		x	x	x	x		x	
MUKANGA*	x	x	x	x	x		x	x	x	x	x	x	x	x	x		x	x	x	x		x	
MULONGO	x	x	x	x	x		x		x			x	x	x	x		x	x	x	x		x	

NID: national immunization day ; SNID: sub-national immunization day; CR- case response
tOPV: trivalent oral polio vaccine; bOPV: bivalent oral polio vaccine; mOPV2: monovalent oral polio vaccine subtype 2
* healthzones which experienced cVDPV2 cases
data source: WHO, * additional source [ALLEMAN]

3.8 References

1. Fine PEM, Carneiro IAM. Transmissibility and Persistence of Oral Polio Vaccine Viruses: Implications for the Global Poliomyelitis Eradication Initiative. *American Journal of Epidemiology.* 1999;150(10):1001-1021.

2. Famulare M, Chang S, Iber J, et al. Sabin Vaccine Reversion in the Field: a Comprehensive Analysis of Sabin-Like Poliovirus Isolates in Nigeria. *Journal of Virology.* 2016;90(1):317-331.

3. Kew O, Morris-Glasgow V, Landaverde M, et al. Outbreak of poliomyelitis in Hispaniola associated with circulating type 1 vaccine-derived poliovirus. *Science.* 2002;296(5566):356-359.

4. Fine PE, Oblapenko G, Sutter RW. Polio control after certification: major issues outstanding. *Bulletin of the World Health Organization.* 2004;82(1):47-52.

5. 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(6489):401-405.

6. Minor P. Vaccine-derived poliovirus (VDPV): Impact on poliomyelitis eradication. *Vaccine.* 2009;27(20):2649-2652.

7. Rousset D, Rakoto-Andrianarivelo M, Razafindratsimandresy R, et al. Recombinant vaccine–derived poliovirus in Madagascar. *Emerging infectious diseases.* 2003;9(7):885.

8. Shimizu H, Thorley B, Paladin FJ, et al. Circulation of type 1 vaccine-derived poliovirus in the Philippines in 2001. *Journal of Virology.* 2004;78(24):13512-13521.

9. Hampton LM, du Châtellier GM, Fournier-Caruana J, et al. Considerations for the Full Global Withdrawal of Oral Polio Vaccine After Eradication of Polio. *The Journal of Infectious Diseases.* 2017;216(Supplement_1):S217-S225.

10. Shahzad A. Time for a Worldwide Shift from Oral Polio Vaccine to Inactivated Polio Vaccine. *Clinical Infectious Diseases.* 2009;49(8):1287-1288.

11. Hull HF, Minor PD. When Can We Stop Using Oral Poliovirus Vaccine? *The Journal of Infectious Diseases.* 2005;192(12):2033-2035.

12. Global Polio Eradication Initiative. *Polio Eradication and Endgame Strategic Plan 2013–2018.* 2014.

13. UNICEF. *cold chain logistics and vaccine management during polio supplementary immunization activities.* 2015.

14. World Health Organization, Global Polio Eradication Initiative. *Polio endgame strategy 2019-2023: eradication, integration, certification and containment.* World Health Organization;2019.

15. Hull HF, Ward NA, Hull BP, Milstien JB, de Quadros C. Paralytic poliomyelitis: seasoned strategies, disappearing disease. *The Lancet.* 1994;343(8909):1331-1337.

16. Alleman MM, Chitale R, Burns CC, et al. Vaccine-derived poliovirus outbreaks and events—three provinces, Democratic Republic of the Congo, 2017. *Morbidity and Mortality Weekly Report.* 2018;67(10):300.

17. Jorba J, Diop OM, Iber J, et al. Update on vaccine-derived polioviruses—worldwide, January 2017–June 2018. *Morbidity and Mortality Weekly Report.* 2018;67(42):1189.

18. Khan F, Datta SD, Quddus A, et al. Progress toward polio eradication—worldwide, January 2016–March 2018. *Morbidity and Mortality Weekly Report.* 2018;67(18):524.

19. Morales M. Progress toward polio eradication—worldwide, 2015–2016. *MMWR Morbidity and mortality weekly report.* 2016;65.

20. Voorman A, Hoff NA, Doshi RH, et al. Polio immunity and the impact of mass immunization campaigns in the Democratic Republic of the Congo. *Vaccine.* 2017;35(42):5693-5699.

21. Deressa W, Kayembe P, Neel AH, Mafuta E, Seme A, Alonge O. Lessons learned from the polio eradication initiative in the Democratic Republic of Congo and Ethiopia: analysis of implementation barriers and strategies. *BMC Public Health.* 2020;20(4):1807.

22. World Health Organization. *Microplanning for immunization service delivery using the Reaching Every District (RED) strategy.* World Health Organization;2009.

23. Halm A, Yalcouyé I, Kamissoko M, et al. Using oral polio vaccine beyond the cold chain: A feasibility study conducted during the national immunization campaign in Mali. *Vaccine.* 2010;28(19):3467-3472.

24. Shrivastava A, Gupta N, Upadhyay P, Puliyel J. Caution needed in using oral polio vaccine beyond the cold chain: vaccine vial monitors may be unreliable at high temperatures. *Indian J Med Res.* 2012;135(4):520-522.

25. Mensi C, Pregliasco F. Poliomyelitis: Present Epidemiological Situation and Vaccination Problems. *Clinical Diagnostic Laboratory Immunology.* 1998;5(3):278-280.

26. MacNeil A, Lee C-w, Dietz V. Issues and considerations in the use of serologic biomarkers for classifying vaccination history in household surveys. *Vaccine.* 2014;32(39):4893-4900.

27. Cutts FT, Izurieta HS, Rhoda DA. Measuring coverage in MNCH: design, implementation, and interpretation challenges associated with tracking vaccination coverage using household surveys. *PLoS Med.* 2013;10(5):e1001404.

28. Halbrook M, Alfonso VH, Hoff NA, et al. Vaccination and serologic response against poliovirus among children in an outbreak area of vaccine derived poliovirus in the Democratic Republic of the Congo. In:2020.

29. Halbrook M, Gadoth A, Hoff N, Weldon WC, W.; RA. Poliovirus Seroprevalence and Vaccine Habits in a Vaccine-Derived Poliovirus Outbreak Site in the Democratic Republic of Congo, 2018. In:2020.

30. Alleman MM, Chitale R, Burns CC, et al. Vaccine-Derived Poliovirus Outbreaks and Events - Three Provinces, Democratic Republic of the Congo, 2017. *MMWR Morbidity and mortality weekly report.* 2018;67(10):300-305.

31. Weldon WC, Oberste MS, Pallansch MA. Standardized Methods for Detection of Poliovirus Antibodies. In: Martín J, ed. *Poliovirus: Methods and Protocols.* New York, NY: Springer New York; 2016:145-176.

32. Hamborsky J, Kroger A. *Epidemiology and prevention of vaccine-preventable diseases, E-Book: The Pink Book.* Public Health Foundation; 2015.

Chapter 4. Spatial Analysis of Vaccine Derived Poliovirus Type 2 Cases and Spatial Risk Factors for Under-immunization, Democratic Republic of the Congo, 2010-2019

4.1 Abstract

Given the persistence of circulating vaccine-derived polio virus type 2 (cVDPV2) in the Democratic Republic of the Congo (DRC) improving the effectiveness of vaccination outreach efforts such as supplementary immunization activities (SIAs) is paramount for polio eradication. Identification of spatial relationships among cVDPV2 cases, vaccine resources and capabilities, and healthcare access can provide a roadmap towards the implementation of SIAs targeted towards communities at higher risk for under-vaccination. This analysis drew upon GIS data from national VDPV2 surveillance, DHS Service Provision Assessment data, and a child vaccine serologic survey in a cVDPV2 outbreak region to identify areas of DRC at greater risk for poor vaccine coverage and VDPV2 emergence. Haut Lomami and Tanganyika provinces remain a region of high likelihood for VDPV2 case emergence, but a high density of cases observed in 2019 outside of this region has now widened the region of elevated cVDPV2 risk. Getis-Ord Gi* hot spot analysis identified spatial clusters where vaccine capabilities were substandard or missing yet many of these cold spots did not overlap cVDPV2 case clusters. However, among participants in the serologic survey, access to health care infrastructure was associated with a presence of neutralizing antibodies. Children who were seropositive lived on average closer to a hospital or a main road compared to children who were not. Planning and implementation of future SIAs can use these spatial relationships to allocated time and resources towards communities with lower rates of vaccination and improve SIA effectiveness and polio vaccine coverage rates.

4.2 Background

Persistent transmission of vaccine-derived polio virus (VDPV) is a key concern for polio eradication, now in its endgame stages.[1,2] Since the withdrawal of poliovirus serotype 2 from routine vaccination several cVDPV2 (circulating VDPV serotype 2) outbreaks have seeded in the Democratic Republic of the Congo (DRC).[3-5] In response to the detection of a new VDPV2 case, the DRC Ministry of Health in conjunction with the World Health Organization (WHO) and the Extended Programme on Immunization (EPI) implements a supplementary immunization activity (SIAs) in affected sub-provincial health administration areas (health zones). During these campaigns, health officials visit every target village and offer polio vaccines door-to-door, most often monovalent oral polio vaccine serotype 2 (mOPV2), to all children under five years of age.[6,7] Despite frequent health outreach activities recorded, polio vaccination coverage in DRC is below necessary herd immunity rates and cVDPV2 cases persist.[8,9]

Vaccination against poliovirus has been instrumental in the success towards eradication however the use of SIAs to improve vaccination coverage has not been without negative impacts. A study in South Africa found SIAs to be associated with a reduction of maternal health services and a multi-country analysis found that SIAs can lead to future healthcare service disruptions.[10,11] Globally, SIAs have been observed to have both positive and negative effects on routine public health services.[12-15] Despite this, improving population immunity against poliovirus through routine childhood immunization and SIAs is currently the primary strategy for VDPV control.

While the DRC EPI reports the timing of all SIAs and the number of vaccine doses administered, the true success of an individual SIA relies on the ability to reach children who have been missed by routine immunization.[16,17] Understanding where under-vaccinated children are, or are likely to

be, remains a challenge in under-resourced areas. The DRC's large land size (2.34 million km^2), poor transportation infrastructure, and variable healthcare record keeping can make finding and accessing under-vaccinated communities a persistent issue.[18] Efforts to measure vaccination coverage, like that of the Demographic and Health Survey (DHS) and smaller scale serologic surveys are resource and time intensive.[19]

Spatial assessments of indicators associated with vaccine coverage can help uncover areas where vaccination coverage is likely to be substandard without the need of participant-based research. Previous research in sub-Saharan Africa has demonstrated that distance to a health facility is associated with delayed or missed childhood vaccinations.[20-28] In addition to proximity measures, the capacities and services available at local health facilities plays a large role in the availability of vaccines for communities. Vaccine cold chain is a system of temperature-controlled transportation and storage of vaccines carefully coordinated in order to maintain potency as vaccines move from manufacturer to individual.[29,30] As short term storage of polio vaccines requires them to be kept at 2°-8° Celsius, the availability of refrigeration and cold chain monitoring at health facilities plays a role in building population immunity against poliovirus.[31-33] Other health facility capabilities, like vaccine stock outs or having necessary staff to offer vaccine outreach to the community likewise has direct impact on local vaccine coverages rates.[34-36] Establishing understandings of how the availability of health care resources impacts childhood vaccine uptake can offer insights into improving future vaccine intervention effectiveness. SIAs are, by design, flexible instruments for vaccine outreach. Quantifying vaccine risk in spatial terms, rather than by socio-demographics or behavioral characteristics can have actionable outcomes for SIA planning and implementation logistics.

This study uses surveillance case data of cVDPV2 cases from 2010-2019, national health facility service provision assessments, and field-based serologic survey data to identify areas of increased risk for VDPV2 emergence and spatial risk factors for under vaccination to poliovirus in the DRC.

4.3 Methods

Data Sources

The geospatial data for this investigation was compiled from several sources, a full list and citations can be found in the appendix. Esri shapefile data for DRC administrative boundaries, roads, and waterways were made available by the Humanitarian Data Exchange, a service managed by the United Nations Office for the Coordination of Humanitarian Affairs; forest raster data was provided open source by the World Resources Institute; and population data at the health zone level was provided by the WorldPop Open Population Repository. National data on DRC health facilities and available health services was obtained from the 2017-18 DHS Service Provision Assessment (SPA).[37,38] SPA sites are selected from comprehensive national lists and are designed to provide indicators for key health service delivery topics including cold chain capability, vaccine storage practices, and vaccination services. Lastly, locations of health facilities in the Haut Lomami and Tanganyika provinces was compiled and made available by the Geo-Referenced Infrastructure and Demographic Data for Development (GRID3) initiative. In addition to publicly available data, spatially referenced VDPV2 case data and information regarding the calendar of SIAs across DRC was made available via collaboration with the DRC EPI.

Finally, we conducted a cross-sectional population-based study in July 2016, and March 2018 across ten health zones in the cVDPV2-affected provinces of Haut Lomami and

Tanganyika to measure population seroimmunity to poliovirus. The village sampling and lab methodology has been described in detail elsewhere[39,40] but briefly, children under five years of age and their parent or guardian from selected villages were invited to enroll and participate in a household survey, a child health survey, and a dried blood spot sample collection via finger or heel prick. Five villages were randomly selected from each health zone such that no two villages fell within the same health area (a sub-health zone administrative unit) or were within 500 meters of each other. Questionnaire data and geolocation was collected with Open Data Kit 2012 version 2.0.4 (UW-CSE, Seattle, WA) on tablets.

Data Analysis

Kernel density estimation is a useful tool for visualizing the density of cases across a space and estimating an intensity function of a point process.[41-43] One key element of kernel density estimation is the selection of the kernel bandwidth. Bandwidths were chosen using a Gaussian kernel density estimator applying Silverman's rule of thumb, defaulting to 0.9 times the minimum of the standard deviation and the interquartile range divided by 1.34 times the sample size to the negative one-fifth power and modified for a two-dimensional space.[44-46] Spatial clustering of VDPV2 cases was assessed via Ripley's *K* function which describes spatial clustering as neighborhood size changes.[47] Theoretically, the *K* function is estimated by the number of extra events within a distance *r* of a randomly chosen event divided by the density of events. Under an assumption of complete spatial randomness (CSR), the *K* function can be estimated as simply $K(r) = \pi r^2$. To account for the shape of the DRC national border and reduce bias, an edge correction was employed. Point density and clustering were assessed using the *spatstat* package for R (R Core Team 2014).[48]

K estimate with Ripley edge correction[47]:

$$\widehat{K}(r) = \lambda^{-1} \sum_{i} \sum_{j \neq 1} w(l_i, l_j)^{-1} \frac{I(d_{ij} < r)}{N}$$

To assess areas with poor polio vaccination services and capabilities four health facility functions were analyzed using the Getis-Ord Gi* statistic (local G-statistic)[49-51] in ArcGIS (Esri, Redlands, CA)[52]: 1) observed cold chain monitoring system; 2) available polio vaccines on the day of observation; 3) routinely storing vaccines; and 4) offering childhood vaccination services. The Gi* statistic produces spatial hot and cold spots by comparing a selected attribute of each point in the context of neighboring features to the entire study area and computes a z-score. This z-score is then used to compare if a point's features are clustered spatially at a significance of α= 0.01, 0.05, and 0.10. Statistically significant positive z-scores correspond to hot spots, or in this analysis, clusters of health facilities where vaccination capabilities are present and more concentrated than the study area at large. Negative significant z-scores correspond to cold spots, health facilities where the absence of a capability or service are spatially clustered. The Getis-Ord Gi* statistic is useful in the context of service provision mapping in a low-resource country like the DRC because it can compare heterogenous neighborhoods to a heterogenous distribution across the study site and determine clusters where services are missing at rates greater than the national average.

Finally, the relationship between access to healthcare resources and an individual's serostatus and their previous interaction with a polio vaccination intervention was analyzed via t-test. Access to health care was measured vis distance to a nearest hospital or health center and distance to nearest main road using the ArcGIS *near* tool. DRC roadways were categorized using the OpenStreetMap ordinal classification system.[53] From that list, a road was considered a main

road if it was classified as a ‘primary’, ‘secondary’, or ‘tertiary’ roadway. After extraction of distances, t-test comparisons were conducted in SAS v9.4 (Cary, NC).

4.4 Results

Since 2010, 180 cases of cVDPV2 and 13 cases of aVDPV2 have been recorded in the DRC. Of these, 164 (91.1%) cases of cVDPV2 have recorded geospatial locations made available by the DRC EPI. cVDPV2 cases have been observed in 48 health zones across 14 provinces—the greatest incidence has been observed in Bena-Diele (n=16, Sankuru province) and Malemba-Nkulu health zone (n=16, Haut Lomami province). Notably, 56 cases (56/164, 34%) have been observed in Haut Lomami province, and all 28 cases recorded in Sankuru province occurred in 2019 (Figure 4.1).

Kernel density estimation of cases across the entire study period found that the highest likelihood of cases was observed in southeastern Haut Lomami province and western Tangankyika. While the study period encompasses a nine-year period, DRC experienced a four-year gap between cVDPV2 cases from 2011-2016. Kernel density estimates from 2010-2012 found the greatest likelihood of cases in southeastern DRC. Estimates of case likelihood using cases from 2017-2019 found a much broader area where likelihood for case emergence was elevated (Figure 4.2).

A plot of the estimated *K* function confirms that cVDPV2 cases are not homogenously distributed and are indeed clustered. The probability of cases being closer together is much higher than would be expected under complete spatial randomness. As clustering of infectious disease is often a function of population density, the *K* function for observed cases was compared not only to a homogenous Poisson process but also to a simulated point process of 164 cases drawn from a population density function of the DRC. Observed cases were also found to be

more clustered than would be expected given the underlying population density of the DRC (Figure 4.3).

The availability of the four health facility vaccination capabilities of interest varied among DHS SPA sites and across the DRC. Among the selected health facilities, 73.8% offered vaccination services to children but 56.5% of facilities reported that they do not routinely store vaccines. Just 23.9% of SPA sites had a cold chain monitoring system; though among the sites that reported regularly storing vaccines 76.3% had a cold chain monitoring system. With regards to polio vaccines specifically, 71.2% of health facilities reported offering polio vaccines at least one day out of the month (μ=4 days, median= 2 days) but just 27.1% of visited facilities had polio vaccines in stock on the day of study observation. Spatial clustering analysis of these vaccination functions found areas across the nation where a lack of service availability and capability was clustered. Hot spots—areas with higher concentrations of vaccination capabilities —and cold spots—areas with lower concentration of vaccine services—varied across the four vaccination functions analyzed (Figure 4.4). However, a consistent hot spot was observed around Lubumbashi, DRC's second largest city in the southeast corner of the nation, and in Maniema province. Interestingly, Kinshasa, the nation's capital and largest city, was only observed to be a hot spot for child vaccination services. Cold spots of cold chain monitoring systems were observed in Mongala and Sud Ubangi provinces in northwest DRC and clusters of health facilities which did not offer child vaccination services were observed in Kwilu and Nord Kivu provinces. Clusters of health facilities which did not have polio vaccines in stock were observed in several health zones where VDPV cases has also been observed including health zones with high VDPV incidence in Sankuru, Haut Lomami, and Tanganyika provinces. However, some hot

spots where health facilities with polio vaccine in stock were clustered was also observed in VDPV-affected health zones.

Among the families with children under five years of age which participated in the serologic survey, distance to the nearest health facility and distance to the nearest main road was associated with seroreactivity to serotype 1, 2, and 3. On average participants lived 27.2 kilometers from a hospital and 2.7 kilometers away from a health center. Children without seroreactivity to serotypes 1 or 3 lived 7.4 and 7.6 kilometers further, respectively, from a hospital than those with seroreactivity (Table 4.1). Children who were not seroreactive to serotype 2 lived, on average, 13.1 kilometers further from a hospital than children with observed neutralizing antibodies to serotype 2. No significant difference in distance to a health center was detected between those with and without neutralizing antibodies (data not shown). Additionally, seroreactivity was associated with the kernel density bandwidth estimated for health facilities—participants that were seroreactive to serotypes 1, 2, and 3 were observed to live in areas with higher health facility density.

Study participants were asked two questions regarding their own participation in a polio vaccination outreach campaign: 1) has someone visited your home to discuss polio vaccination and 2) has someone come to your home offering polio vaccinations. On average, participants who answered ‘yes’ to either of these questions lived around 1.5 km closer a main road compared to those who answered ‘no’. Participants who answered yes to having someone offering a polio vaccine visit their home lived 6.6 km closer to a hospital compared to those who did not.

4.5 Discussion

Review of the 164 cVDPV cases from 2010-2019 for which spatial coordinates were available confirms that the health zones along the border of Haut Lomami and Tanganyika province has the highest likelihood for case emergence. Observed cases were found to be clustered compared to both a homogenous Poison distribution representing total spatial randomness and a distribution of cases modeled using an underlying spatial distribution derived from the population density of the DRC. Interestingly, areas which had a higher density of cVDPV2 cases did not overlap with high population areas. As much of the population of DRC is concentrated in two major cities—Kinshasa in the west and Lubumbashi in the southeast—the absence of cVDPV2 cases in these areas perhaps reflects the vaccine resource imbalance that exists between urban and rural areas in DRC and the persistence of cVDPV2 transmission following nearly a decade of cases. Of note, Kinshasa did experience one cVDPV2 case in 2019 and five in 2020.

Indeed, key healthcare resources that are critical in polio vaccination were not observed to be equally distributed across DRC. Identifying areas with poor vaccination services is crucial to achieving geographic parity of vaccine distribution. Interestingly, few common cold spots were found across the four vaccination capacities investigated and locations which did have large clusters of health facilities lacking key vaccine capabilities—like the cluster in Ituri province of facilities without cold chain capability or the cluster in eastern Mongala of facilities which did not offer child vaccination services—were not sites of large cVDPV2 clusters. In the health zones along the Haut Lomami-Tanganyika border where the greatest case intensity was observed there were health facilities that did not have polio vaccines in stock clustered with 90% and 95% confidence. Taken together, understanding where cVDPV2 cases are clustered and where

vaccination capabilities are subpar can be used to identify areas where vaccine coverage is likely below target levels. Yet, this cluster analysis cannot fully explain why Haut Lomami and Tanganyika provinces have repeatedly experienced the most cases as their vaccine services were fairly consistent with national rates of service provision. Additionally, an estimation of population seroprevalence to poliovirus collected during the 2014 DHS did not find seroprevalence in this region (former Katanga province) to be significantly different from the rest of the nation.[54] Further investigation is needed to understand why this area is uniquely affected by cVDPV2.

One possible explanation is that the frequent use of SIAs in cVDPV2 endemic regions may itself increase the risk of VDPV emergence by creating chronically over-vaccinated and consistently missed groups of children. A modelling study of VDPV risk in Nigeria found that as routine immunization coverage rates drop, just one SIA may increase the risk of VDPV emergence and that while up to three SIAs may be needed to reduce VDPV risk, there existed a threshold where additional SIAs no longer contributed to VDPV control.[55,56]

Today, SIA planning in DRC relies on a mix of written microplans, an operational planning document which contains population and village lists, and satellite imagery to confirm settlement locations.[57] While the incorporation of GIS mapping has improved SIA planning, SIAs still aim for widespread coverage, when perhaps, a targeted methodology might be more effective at closing the immunity gap.[58] On average, participants in our serologic survey who did not have detectable antibodies to poliovirus, indicating a likely lack of vaccination, lived further away from a hospital or a main road compared to those that did have a detectable presence of antibodies. Distance to a health center was not associated with polio serostatus. However, since health facilities can have variable resources available, proximity to a singular health center may

not be a reliable indicator of proximity to actual healthcare services. When poliovirus serostatus of a participant was analyzed in comparison to the density of health centers in their area, presence of poliovirus antibodies was associated with increased health facility density. As SIAs work to close the immunity gap, prioritizing high risk communities, like those located in clusters lacking vaccine capacities or communities far from health infrastructure, can improve SIA performance.

This study utilizes surveillance and population data recorded across a nine-year period. As such variability could have been introduced as surveillance strategies and record keeping practices evolved. Additionally, most VDPV cases are identified when an individual presents with acute flaccid paralysis (AFP), the most well-known symptom of poliovirus, but one that has poor sensitivity with a <1% incidence rate among infected children. As such, known VDPV cases may represent just 1% of all infections. Additionally, as AFP has multiple etiologies, laboratory identification is necessary prior to confirming a VDPV cases.[59] This process can lead to a miscount of cases and missing data issues as several pieces of information are required in order to confirm a VDPV case. Another limitation can arise from aggregating information at the health zone level. While point process methods can overcome this, the use of health zone level population data smoothed at the centroid to estimate the population density could have led to misrepresentations of DRC's population. Finally, this analysis was spatially constrained to the DRC border yet, from 2006-2011 wildtype poliovirus was imported into DRC from neighboring Angola several times and some cVDPV2 strains observed in DRC have been genetically linked to Angola.[60,61] An analysis of VDPV risk which looks more broadly at the central Africa region may provide necessary context for how VDPV clusters arise.

Overall, emergence of cVDPV2 cases in DRC since 2010 has been clustered in several regions. While vaccination is the primary strategy for cVDPV2 control, overuse of SIAs, especially if SIAs create chronically missed pockets of the population, may exacerbate VDPV2 spread.[3,62] Spatial barriers to access like distance to a hospital or ease of transport to high functioning health facilities is associated with under-vaccination. Planning and implementations of future SIAs can use these spatial relationships to allocated time and resources towards communities with lower rates of vaccination and improve SIA effectiveness. Further research into why and how cVDPV2 clusters arise in DRC where they do would provide meaningful context into future control methods and interventions.

4.6 Tables and Figures

Figure 4.1. cVDPV2 cases, 2010-2019

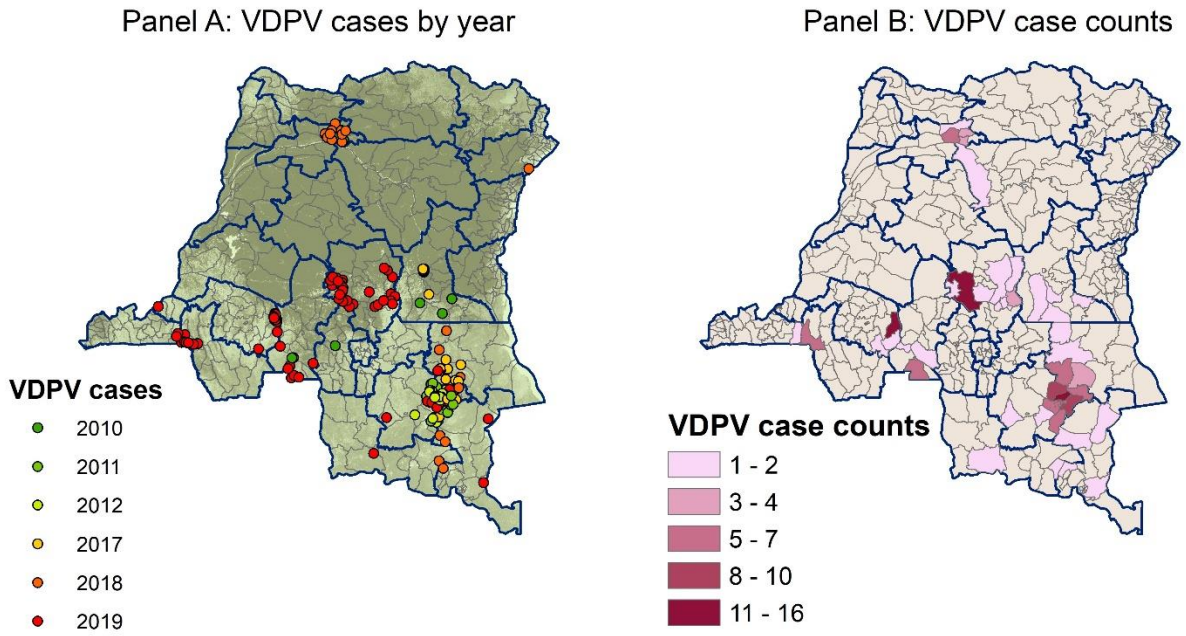


Figure 4.2. Intensity of cVDPV2 cases

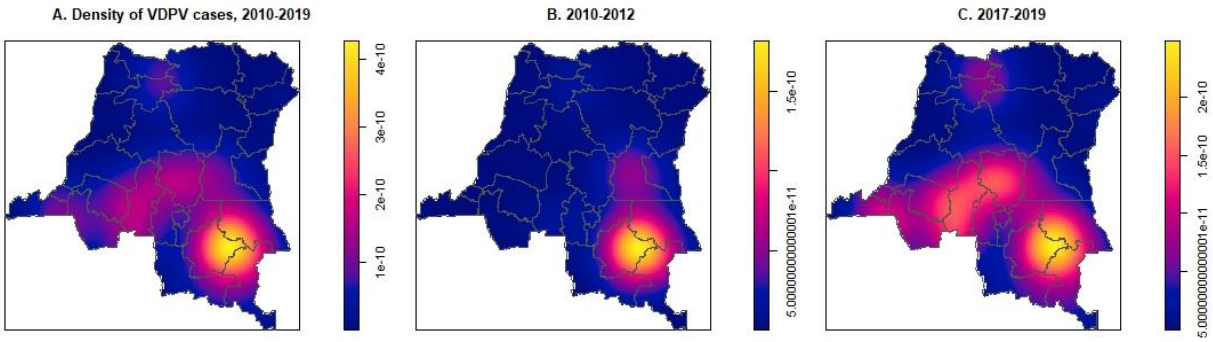


Figure 4.3. Simulated cVDPV2 cases and cluster analysis

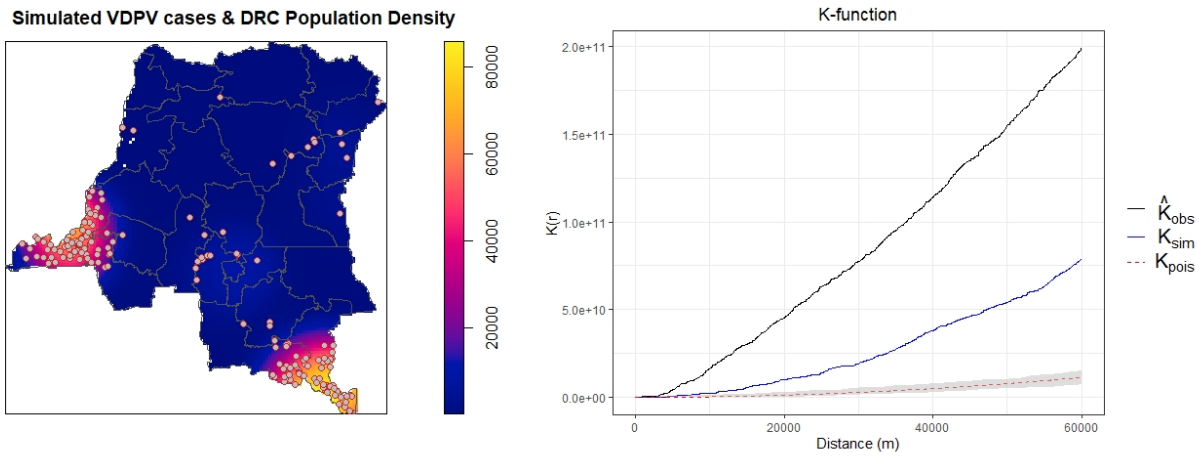


Figure 4.4. Hot Spot Analysis of DHS SPA Site Vaccination Capacity

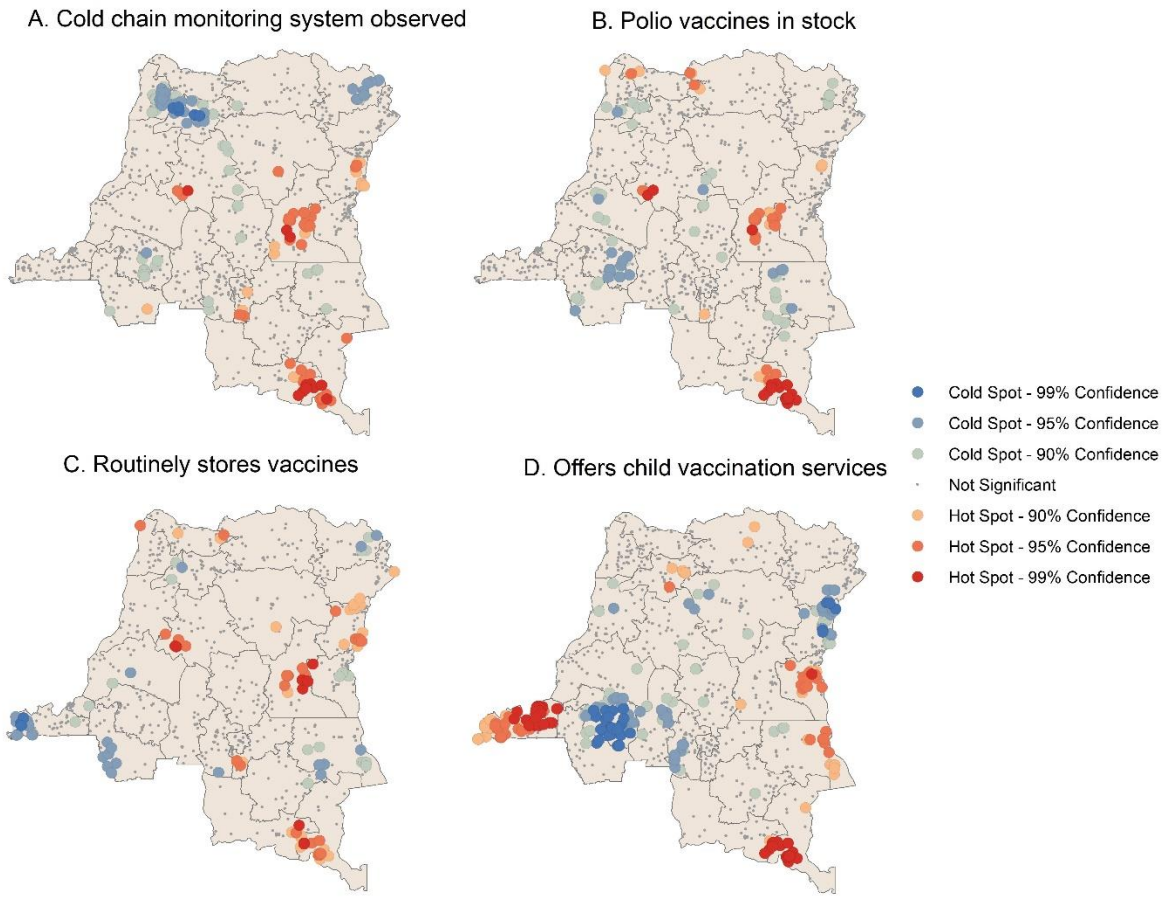


Table 4.1. Relationship between poliovirus serology and vaccine outreach and access to healthcare, measured by kilometers

Distance (km) to nearest:	Reactivity to polio serotype						Polio vaccination outreach			
	Type 1		Type 2		Type 3		Home visit		Home vaccine	
	km	p-value	km	p-value	km	p-value	km	p-value	km	p-value
Hospital	-7.38	<.0001	-13.1	<.0001	-7.63	<.0001	-1.57	0.3986	-6.56	0.0007
Main road	-0.45	0.2473	-1.54	0.0002	-0.79	0.0368	-1.53	0.0052	-1.66	0.0008

Comparing distance among those seroreactive to poliovirus compared to those seronegative and those who answered 'yes' to having a health care worker visit their home to share information and administer a vaccine compared to those who answered 'no'.

4.7 Appendix

Appendix Table 4.1. GIS data sources

Map Layer	**Source**
DRC Administrative Boundaries	Humanitarian Data Exchange[63]
Forest Raster data	World Resources Institute- datasets.wri.org
Roads	Humanitarian OpenStreetMap Team (HOT)[64]
Health Facility data	GRID3[65]
VDPV cases	DRC EPI Surveillance Data
population denominators	WorldPop Open Population Repository[66,67]

4.8 References

1. Global Polio Eradication Initiative. *Polio Eradication and Endgame Strategic Plan 2013–2018.* 2014.

2. World Health Organization, Global Polio Eradication Initiative. *Polio endgame strategy 2019-2023: eradication, integration, certification and containment.* World Health Organization;2019.

3. 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(6489):401-405.

4. Mbaeyi C, Alleman MM, Ehrhardt D, et al. Update on Vaccine-Derived Poliovirus Outbreaks - Democratic Republic of the Congo and Horn of Africa, 2017-2018. *MMWR Morbidity and mortality weekly report.* 2019;68(9):225-230.

5. Alleman MM, Chitale R, Burns CC, et al. Vaccine-derived poliovirus outbreaks and events—three provinces, Democratic Republic of the Congo, 2017. *Morbidity and Mortality Weekly Report.* 2018;67(10):300.

6. Global Polio Eradication Initiative. mOPV2 Management, Monitoring, Removal and Disposal. In:2021.

7. Global Polio Eradication Initiative. *Standard Operating Procedures: Responding to a Poliovirus Event or Outbreak.* 2019.

8. Declining vaccination rates in the Democratic Republic of the Congo could lead to resurgence in deadly diseases [press release]. unicef.org2020.

9. Kew OM, Sutter RW, Gourville EMd, Dowdle WR, Pallansch MA. Vaccine-Derived Polioviruses and the Endgame Strategy for Global Polio Eradication. *Annual Review of Microbiology.* 2005;59(1):587-635.

10. Verguet S, Jassat W, Bertram MY, et al. Impact of supplemental immunisation activity (SIA) campaigns on health systems: findings from South Africa. *Journal of Epidemiology and Community Health.* 2013;67(11):947-952.

11. Closser S, Cox K, Parris TM, et al. The impact of polio eradication on routine immunization and primary health care: a mixed-methods study. *The Journal of infectious diseases.* 2014;210(suppl_1):S504-S513.

12. Clements CJ, Soakai TS, Sadr-Azodi N. A review of measles supplementary immunization activities and the implications for Pacific Island countries and territories. *Expert Review of Vaccines.* 2017;16(2):161-174.

13. Helleringer S, Asuming PO, Abdelwahab J. The effect of mass vaccination campaigns against polio on the utilization of routine immunization services: A regression discontinuity design. *Vaccine.* 2016;34(33):3817-3822.

14. Haenssgen MJ. Impact of high-intensity polio eradication activities on children's routine immunization status in Northern India. *Health Policy and Planning.* 2017;32(6):800-808.

15. Chakrabarti A, Grépin KA, Helleringer S. The impact of supplementary immunization activities on routine vaccination coverage: An instrumental variable analysis in five low-income countries. *PLOS ONE.* 2019;14(2):e0212049.

16. World Health Organization. Planning and implementing high-quality supplementary immunization activities for injectable vaccines using an example of measles and rubella vaccines: field guide. 2016.

17. Hsu CH, Kader M, Mahamud A, et al. Progress toward poliomyelitis eradication—Pakistan, January 2018–September 2019. *Morbidity and Mortality Weekly Report.* 2019;68(45):1029.

18. Ashbaugh HR, Hoff NA, Doshi RH, et al. Predictors of measles vaccination coverage among children 6–59 months of age in the Democratic Republic of the Congo. *Vaccine.* 2018;36(4):587-593.

19. Shenton LM, Wagner AL, Ji M, Carlson BF, Boulton ML. Vaccination assessments using the Demographic and Health Survey, 2005-2018: a scoping review. *BMJ Open.* 2020;10(12):e039693.

20. Wiysonge CS, Uthman OA, Ndumbe PM, Hussey GD. Individual and contextual factors associated with low childhood immunisation coverage in sub-Saharan Africa: a multilevel analysis. *PloS one.* 2012;7(5):e37905.

21. Hajizadeh M. Socioeconomic inequalities in child vaccination in low/middle-income countries: what accounts for the differences? *J Epidemiol Community Health.* 2018;72(8):719-725.

22. Okwaraji YB, Mulholland K, Schellenberg J, Andarge G, Admassu M, Edmond KM. The association between travel time to health facilities and childhood vaccine coverage in rural Ethiopia. A community based cross sectional study. *BMC public health.* 2012;12(1):1-9.

23. Vyas P, Kim D, Adams A. Understanding spatial and contextual factors influencing intraregional differences in child vaccination coverage in Bangladesh. *Asia Pacific Journal of Public Health.* 2019;31(1):51-60.

24. Shemwell SA, Peratikos MB, Gonzalez-Calvo L, et al. Determinants of full vaccination status in children aged 12–23 months in Gurùé and Milange districts, Mozambique: results of a population-based cross-sectional survey. *International health.* 2017;9(4):234-242.

25. Legesse E, Dechasa W. An assessment of child immunization coverage and its determinants in Sinana District, Southeast Ethiopia. *BMC pediatrics.* 2015;15(1):1-14.

26. Jani JV, De Schacht C, Jani IV, Bjune G. Risk factors for incomplete vaccination and missed opportunity for immunization in rural Mozambique. *BMC public health.* 2008;8(1):1-7.

27. Sato R. Association between access to a health facility and continuum of vaccination behaviors among Nigerian children. *Hum Vaccin Immunother.* 2020;16(5):1215-1220.

28. Blanford JI, Kumar S, Luo W, MacEachren AM. It's a long, long walk: accessibility to hospitals, maternity and integrated health centers in Niger. *International Journal of Health Geographics.* 2012;11(1):24.

29. World Health Organization. WHO: Global Programme For Vaccines And Immunization Expanded Programme On Immunization. *Immunity and vaccines in safe vaccine handling, cold chain and immunizations World Health Organization, Geneva.* 1998:44-208.

30. Yauba S, Harmelle EE, Marius VZ, et al. Availability and Status of Vaccine Cold Chain Equipment in Cameroon. *Journal of Vaccines & Vaccination.* 2019;10(2):1-6.

31. Bankole AM, Olusegun K-K, Marian NB, et al. The impact of health facility monitoring on cold chain management practices in Lagos, Nigeria. *Journal of Public Health and Epidemiology.* 2010;2(4):78-81.

32. Ateudjieu J, Kenfack B, Nkontchou BW, Demanou M. Program on immunization and cold chain monitoring: the status in eight health districts in Cameroon. *BMC research notes.* 2013;6(1):1-7.

33. Sokhey J, Gupta CK, Sharma B, Singh H. Stability of oral polio vaccine at different temperatures. *Vaccine.* 1988;6(1):12-13.

34. Burnett RJ, Mmoledi G, Ngcobo NJ, Dochez C, Seheri LM, Mphahlele MJ. Impact of vaccine stock-outs on infant vaccination coverage: a hospital-based survey from South Africa. *International Health.* 2018;10(5):376-381.

35. Iwu CJ, Ngcobo N, Jaca A, et al. A systematic review of vaccine availability at the national, district, and health facility level in the WHO African Region. *Expert Review of Vaccines.* 2020;19(7):639-651.

36. Sato R. Vaccine stockouts and vaccination rate: examination of unique administrative data from Nigeria. *International health.* 2020;12(3):170-176.

37. Ecole de Sante Publique de Kinshasa. Republique Democratique du Congo Evaluation des Presentations des Services de souns de Sante EPSS 2017-2018 Rapport Final [Democratic Rebublic of Congo Evaluation of Health Services EPSS 2017-2018 Final report], 2019. *SPA30 pdf.*

38. The Demographic and Health Program. Service Provision Assessments (SPA). https://dhsprogram.com/Methodology/Survey-Types/SPA.cfm, 2021.

39. Halbrook M, Alfonso VH, Hoff NA, et al. Vaccination and serologic response against poliovirus among children in an outbreak area of vaccine derived poliovirus in the Democratic Republic of the Congo. In:2020.

40. Halbrook M, Gadoth A, Hoff N, Weldon WC, W.; RA. Poliovirus Seroprevalence and Vaccine Habits in a Vaccine-Derived Poliovirus Outbreak Site in the Democratic Republic of Congo, 2018. In:2020.

41. Shi X. Selection of bandwidth type and adjustment side in kernel density estimation over inhomogeneous backgrounds. *International Journal of Geographical Information Science.* 2010;24(5):643-660.

42. Baddeley A, Rubak E, Turner R. *Spatial point patterns: methodology and applications with R.* CRC press; 2015.

43. Diggle P. Statistical analysis of spatial point patterns, London: Arnold. *MATH Google Scholar.* 2003.

44. Gramacki A. Bandwidth selectors for kernel density estimation. In: *Nonparametric Kernel Density Estimation and Its Computational Aspects.* Springer; 2018:63-83.

45. Sheather SJ, Jones MC. A reliable data-based bandwidth selection method for kernel density estimation. *Journal of the Royal Statistical Society: Series B (Methodological).* 1991;53(3):683-690.

46. Scott DW. *Multivariate density estimation: theory, practice, and visualization.* John Wiley & Sons; 2015.

47. Dixon PM. Ripley's K function. *Wiley StatsRef: Statistics Reference Online.* 2014.

48. Baddeley AJ, Turner R. Spatstat: An R package for analyzing spatial point pattens. In: University of Western Australia. Department of Mathematics and Statistics; 2004.

49. Getis A, Ord JK. The analysis of spatial association by use of distance statistics. In: *Perspectives on spatial data analysis.* Springer; 2010:127-145.

50. Murad A, Khashoggi BF. Using GIS for disease mapping and clustering in Jeddah, Saudi Arabia. *ISPRS International Journal of Geo-Information.* 2020;9(5):328.

51. Zulu LC, Kalipeni E, Johannes E. Analyzing spatial clustering and the spatiotemporal nature and trends of HIV/AIDS prevalence using GIS: the case of Malawi, 1994-2010. *BMC infectious diseases.* 2014;14(1):1-21.

52. Mitchel A. *The ESRI Guide to GIS analysis, Volume 2: Spartial measurements and statistics.* ESRI press; 2005.

53. wiki.Openstreetmap.org. Key:highway. https://wiki.openstreetmap.org/wiki/Key:highway.

54. Voorman A, Hoff NA, Doshi RH, et al. Polio immunity and the impact of mass immunization campaigns in the Democratic Republic of the Congo. *Vaccine.* 2017;35(42):5693-5699.

55. Pons-Salort M, Burns CC, Lyons H, et al. Preventing Vaccine-Derived Poliovirus Emergence during the Polio Endgame. *PLOS Pathogens.* 2016;12(7):e1005728.

56. Duintjer Tebbens RJ, Pallansch MA, Wassilak SGF, Cochi SL, Thompson KM. Combinations of Quality and Frequency of Immunization Activities to Stop and Prevent Poliovirus Transmission in the High-Risk Area of Northwest Nigeria. *PLOS ONE.* 2015;10(6):e0130123.

57. World Health Organization. *Microplanning for immunization service delivery using the Reaching Every District (RED) strategy.* World Health Organization;2009.

58. Gammino VM, Nuhu A, Chenoweth P, et al. Using geographic information systems to track polio vaccination team performance: pilot project report. *The Journal of infectious diseases.* 2014;210(suppl_1):S98-S101.

59. Andrus JK, De Quadros C, Olivé JM, Hull HF. Screening of cases of acute flaccid paralysis for poliomyelitis eradication: ways to improve specificity. *Bulletin of the World Health Organization.* 1992;70(5):591.

60. Gumede N, Lentsoane O, Burns CC, et al. Emergence of Vaccine-derived Polioviruses, Democratic Republic of Congo, 2004–2011. *Emerging Infectious Diseases.* 2013;19(10):1583-1589.

61. Alleman MM, Jorba J, Greene SA, et al. Update on vaccine-derived poliovirus outbreaks—worldwide, July 2019–February 2020. *Morbidity and Mortality Weekly Report.* 2020;69(16):489.

62. McCarthy KA, Chabot-Couture G, Famulare M, Lyons HM, Mercer LD. The risk of type 2 oral polio vaccine use in post-cessation outbreak response. *BMC medicine.* 2017;15(1):1-11.

63. DSNIS. DRC Health Zone and Health Area boundaries. In: Cross AR, ed2021.

64. OpenStreetMap contributors. HOTOSM Democratic Republic of the Congo Roads (OpenStreetMap Export). In: (HOT) HOT, ed2020.

65. Ministère de la Santé Publique République Démocratique du Congo. GRID3 DRC Haut-Lomami and Tanganyika Health Facilities, Version 01 (Beta). In: (GRID3) G-RIaDDfD, ed. Palisades NY2020.

66. WorldPop. DRC population esitmates. In: WorldPop, ed2020.

67. Linard C, Gilbert M, Snow RW, Noor AM, Tatem AJ. Population Distribution, Settlement Patterns and Accessibility across Africa in 2010. *PLOS ONE.* 2012;7(2):e31743.

Chapter 5. Conclusion and Implications

Vaccination against poliovirus has been one of the most successful public health vaccination campaigns in history. In a lifetime, wild poliovirus has been nearly eliminated—currently wild poliovirus type 1 circulates in just two nations and wild types 2 and 3 have been declared eradicated. These successes have made true poliovirus eradication a near possibility, but endgame eradication strategies face an important paradox: as poliovirus strains are prone to recombination, use of the oral polio vaccine (OPV) itself in under-immunized populations can cause vaccine-derived poliovirus (VDPV) infection of pathogenic nature. Complicating this, has been the WHO's decision to remove poliovirus serotype 2 from the OPV used in routine immunization. Following this global switch day, some VDPVs were expected, yet the current landscape of enduring VDPV outbreaks and pockets of waning poliovirus population immunity five years later has been unexpected. In response to this paradox a novel OPV (nOPV) vaccine has been developed in which the Sabin vaccine strain has been adapted to be more resistant to recombination.

In the Democratic Republic of the Congo (DRC), poverty, poor infrastructure, high biodiversity, and governance issues has hindered the control of many vaccine-preventable diseases. This dissertation describes the current state of poliovirus population immunity in a VDPV-outbreak region and the impact supplementary vaccination campaigns (SIAs) have on VDPV control and population immunity.

These studies highlight the difficult nature of participant-based work in a low-resource sub-Saharan African nation. Both the logistics of conducting field research and the resulting methodological challenges and the effectiveness of SIAs are impacted by the difficult to navigate rural terrain of the DRC and the poor quality of maps and microplans. Communities that live

farther away from health resources and transportation infrastructure are less likely to be reached by health interventions. However, once these communities are reached the impact of an individual SIA on population immunity appears to be poor. Repeated efforts in Haut Lomami and Tanganyika province to improve vaccine coverage have not succeeded and as serocoverage rates decrease over time this region remains a hotspot for VDPV cases. Currently, the novel coronavirus-19 pandemic (COVID-19) has further disrupted routine childhood immunizations due to both diminished contact with health systems and national deprioritization of immunization activities in the face of an emergent disease threat. As the primary control method for SARS-CoV-2 is vaccination, lessons learned from previous vaccine preventable disease control interventions and vaccination campaigns will be paramount for the control of the COVID-19 pandemic.

Introduction of the nOPV into this region will likely improve VDPV control, but future immunization interventions, including COVID-19 vaccine strategies, should aim to improve effectiveness through targeting communities most likely to be under-vaccinated and avoiding over-vaccinating already protected individuals. Additionally, while door-to-door vaccination campaigns are necessary, polio vaccine interventions can benefit from a wider scope of attention. Improving health facilities resources, by eliminating vaccine stock-outs, improving cold chain monitoring and community connectivity to healthcare through traditional infrastructure improvements, and poverty reduction measures will likewise improve poliovirus vaccination coverage. Due to the size and resource constraints of the DRC, the healthcare system is largely decentralized. In practice, communities are organized more around local leadership rather than top-down, centralized federal government systems. Health intervention systems, like the structure and planning behind the supplementary immunization campaign can fall short when

they apply a singular national methodology to a nation of individual regions, villages, tribes, and ethnic groups. Understanding risk factors that drive under-immunization in children can help bolster vaccine intervention effectiveness by offering tools for customizing and targeting interventions for specific regions or groups. Vaccines remain the greatest method for controlling polio and as our knowledge of poliovirus epidemiology expands to include endgame eradication dynamics, so too should intervention strategies.