International Journal of Infectious Diseases 83 (2019) 40–43

Contents lists available at ScienceDirect

International Journal of Infectious Diseases

journal homepage: www.elsevier.com/locate/ijid

Case Report

First report of a persistent oropharyngeal infection of type 2 vaccine-derived poliovirus (iVDPV2) in a primary immune deficient (PID) patient after eradication of wild type 2 poliovirus

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ARTICLE INFO

Article history:
Received 13 February 2019
Received in revised form 28 March 2019
Accepted 28 March 2019
Corresponding Editor: Eskild Petersen, Aarhus, Denmark

Keywords:
Primary immune deficiency (PID)
Type 2 vaccine-derived poliovirus (iVDPV2)
Stools
Oropharyngeal mucosa
Contact isolation
Droplet isolation

ABSTRACT

This is the first report of persistent oropharyngeal mucosal infection with type 2 poliovirus (iVDPV2) in a primary immune deficient patient (PID) after wild type 2 poliovirus eradication. The iVDPV2 also established persistence in the gut. iVDPV2 at both loci evolved independently. Persistent oral infections present a potential risk for oral-oral as well as fecal-oral poliovirus transmission during transition to a poliovirus 2-free world.

Introduction

Wild type 2 poliovirus (WPV2) was declared eradicated in September 2015 (Jorba et al., 2018; GPEI, 2015). In April, 2016, the type-2 strain of oral polio vaccine (OPV2) was withdrawn globally from routine use in live vaccines to prevent reemergence of type 2 vaccine derived poliovirus (VDPV2) and vaccine-associated paralytic polio (VAPP) (WHO, 2015). VDPVs behave like wild polioviruses and can cause outbreaks of poliomyelitis (Jenkins et al., 2010). Prolonged excretion of poliovirus by primary immune deficient (PID) patients is rare (Li et al., 2014; Aghamohammadi et al., 2017; Macklin et al., 2017). VDPV2s excreted by immune deficient individuals are a potential source for reemergence after global WPV2 eradication (Aghamohammadi et al., 2017; Alexander et al., 2009).

We present a case study of a child with PID who had separate persistent (≥6 months excretion (Khetsuriani et al., 2003)) PV2 infections in oropharyngeal and gut mucosa. Such dual infections present a potential risk for oral-oral as well as fecal–oral poliovirus transmission. This case demonstrates the importance of identifying persistent oropharyngeal infections and then taking appropriate measures to reduce the risk for person-to-person transmission, especially as we transition into a poliovirus type 2 free world.

Case report

The non-Israeli PID patient was the fourth child of a consanguineous marriage with significant history suggestive of

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https://doi.org/10.1016/j.ijid.2019.03.037

congenital immune deficiency. The patient born on February 20, 2015, was admitted to Sheba Medical Center at 8 months of age due to recurrent infections, failure to thrive (FTT) and spastic monoplegia of his left leg. MRI revealed cystic changes in the basal ganglia and white matter. Immunologic flow cytometric lymphocyte immune phenotyping revealed absence of HLA DR molecules suggesting MHC class II deficiency SCID ("bare lymphocyte syndrome"). A homozygous frame shift mutation in exon 11 of the RFX5 gene, identified by Sanger sequencing confirmed MHC class II SCID. The child was placed in reverse isolation, received prophylactic antimicrobial treatment, and monthly intravenous immunoglobulin (IVIG) treatments. Hematopoietic stem cell transplantation was ruled out due to severe neurologic impairment, continued FTT despite all supportive measures and prophylactic treatment, social issues in the family, and prediction of less than a 60–70% success even with a matching donor (Hanna and Etzioni, 2014). The patient's physical and mental state deteriorated. At the time of death at 2.5 years of age, the patient was receiving analgesics, suffered from hypoalbuminemia, and had diarrhea that was RT-PCR positive for norovirus as well as poliovirus.

A chance finding of prolonged excretion of iVDPV2 in a previous PID patient presenting with diarrhea (Weil et al., 2016) initiated our policy to test stools for poliovirus from all newly admitted pediatric patients presenting with suspected PID at the Sheba Medical Center. A stool sample from the current case was poliovirus-positive. The patient's medical history revealed that the child received trivalent OPV when 2 months old (February, 2016) in accordance with routine immunization schedules at the child's place of birth. Molecular analysis indicated that the isolate was a type 2 poliovirus. Sequence analysis, performed as previously described (Weil et al., 2016; Shulman et al., 2014) revealed that the viral capsid protein 1 of the poliovirus isolate had diverged from Sabin 2 by at least 0.8% (Figure 1) classifying it as an iVDPV2. Both neurovirulence attenuation sites (nucleotide 481 in the 5′UTR and amino acid 143 in VP1) had reverted to the neurovirulent genotype.

The previously identified PID patient with a persistent iVDPV2 infection was still hospitalized in the same ward as the current case. Therefore our first concern was to determine whether iVDPV2 transmission had occurred within the PID ward. The VP1 sequences of isolates from the two patients were genetically unrelated, ruling out transmission between the patients.

CSF and a throat swab from the patient, and stools from ten contacts, four healthcare workers and six family members from the community, were tested for iVDPV2 immediately after identification of the first VDPV2-positive stool. CSF from the patient and stools from contacts were negative by RT-PCR for poliovirus and

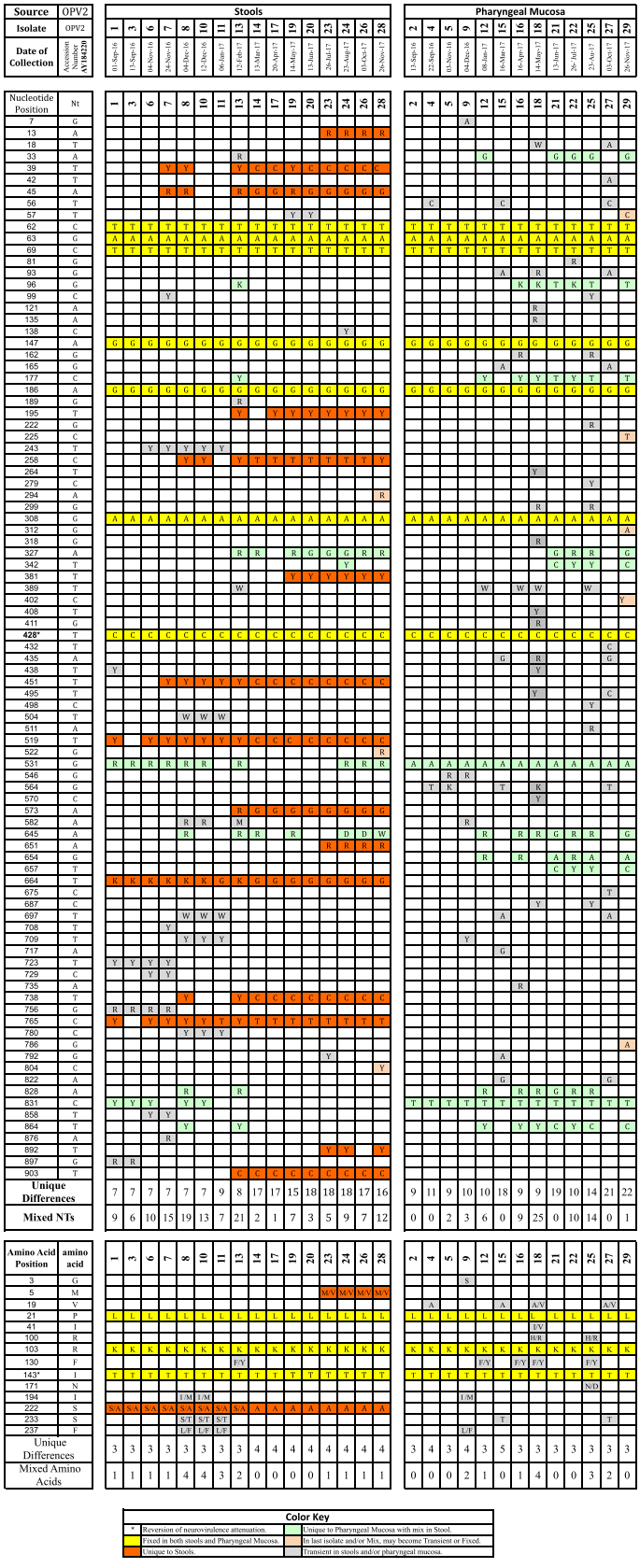

Figure 1. Nucleotide and amino acid substitutions in vaccine-derived type-two poliovirus isolated from stools and pharyngeal mucosa during the prolonged infection of a PID case.

RNA was extracted from virus isolations of stool suspensions and throat swabs, as described previously (; Shulman et al., 2014). Forward primer Y7R and reverse primer Q8 were used to amplify and sequence the viral capsid protein 1 on an ABI 3500 Genetic Analyzer (Applied Biosystems, Foster City, CA, United States) using an ABI PRISM BigDye Terminator Cycle Sequencing kit (Applied Biosystems) as described (Weil et al., 2016; Shulman et al., 2014). The VP1 sequences were compared using the Sequencher v5.2.2 program (GenCodes, Anne Arbor, MI, USA). Nucleotide positions where sequence analysis indicated the presence of a mixture of viruses with different nucleotides at that position are indicated by the degenerate IUB nucleotide code. Two amino acids separated by a "/" indicate amino acid positions where the codon contained a nucleotide position with a degenerate nucleotide and where *in silico* translation of the codon (using Sequencher) depended on which nucleotide was chosen for translation.

Key for highlighting: yellow indicates nucleotide or amino acid substitutions from Sabin 2 (**AY184220**) fixed in all iVDPVs; orange indicates those that emerged and became fixed and were unique for stool isolates; green those that emerged and became fixed and were unique for oropharyngeal isolates although they may appear as minor components in stools; grey indicates a transient substitution; and light pink indicates a substitution in the last isolate that may become fixed or be transient. An "*" indicates nucleotide and amino acid substitutions that reverse attenuation of neurovirulence. The viral capsid protein 1 sequences of iVDPV2 isolates 1–29 were assigned GenBank accession numbers **MK660464**-**MK660492**, respectively.

non-polio enteroviruses, however the PID's throat swab was positive for iVDPV2. No further testing of contacts was performed. We continued to periodically test stools and throat swabs on the dates indicated in Figure 1. All stool and throat swab samples remained iVDPV2-positive until the patient died 21 months after the first poliovirus-positive stool sample.

VP1 nucleotide and amino acid differences from Sabin 2 (GenBank accession number **AY184220**) are presented for sixteen stool and thirteen throat samples spanning the first 14 months of persistent excretion after the initial iVDPV2-positive stool sample (Figure 1). Fixed substitutions are indicated by standard nucleotide base codes A, T, G, or C, whereas nucleotide mixtures indicating two populations of iVDPV2s are indicated by IUB codes, K (G or T), M (A or C), R (A or G), W (A or T), and Y (C and T).

During the 14-month interval, iVDPV2 isolates continued to diverge from Sabin 2 as seen from the kinetic pattern of nucleotide and amino acid substitutions in the iVDPV2 isolates. Seven nucleotides and three amino acid substitutions were present in all isolates (highlighted in yellow) demonstrating a common initial evolutionary pathway. There are unique fixed and mixed substitutions in iVDPV2s isolated from stools (boxes highlighted in red). Within 6 months from the first isolation, ten substitutions became fixed exclusively in iVDPV2s isolated from stools. Within 14 months from the first isolation, another five substitutions emerged in some of the isolates from stool. These fixed and mixed substitutions were not observed in iVDPV2s isolated from throat swabs. Different fixed and mixed substitutions were observed in iVDPV2s isolated from throat swabs but with traces in stools (boxes highlighted in green). This kinetic pattern of nucleotide and amino acid substitutions supports subsequent independent evolution for separate persistent co-infections of iVDPV2 in the oropharyngeal mucosa and the gut of the patient.

During the persistent iVDPV2 co-infections the patient was intermittently discharged and re-admitted. While hospitalized, the PID patient was kept in strict droplet isolation for infection control. Continued droplet isolation was justified by the isolation of iVDPV2 from the throat swab. Parents were instructed to continue droplet and contact isolation while the patient was at home.

Discussion

One of the main tasks of the Global Poliovirus Laboratory Network (GPLN) for the Global Poliovirus Eradication Initiative (GPEI), is to diagnose active poliovirus infections (Jorba et al., 2018; WHO, 2004). Poliovirus infections of gut mucosa last much longer than infections of oropharyngeal mucosa (Alexander et al., 1997; WHO, 2004). Since poliovirus isolation from stools offered the highest chance for successfully isolating live poliovirus, it was selected as the diagnostic method of choice for investigating suspected cases of poliovirus infection (WHO, 2004). Testing throat swabs for poliovirus is not usually performed. There are only a few reports showing evidence for persistent poliovirus from throat swabs from PID patients (Bellmunt et al., 1999; Yang et al., 2005). In one of these reports, (Bellmunt et al., 1999), genomic analysis of the iVDPV1s isolates from the oropharyngeal mucosa and from the gut demonstrated that genetically related iVDPVs had established persistence at both loci and subsequently evolved separately, as we report here. iVDPV1s were periodically isolated over the next 6 years from 43 stool collections. No additional analyses of oropharyngeal mucosa were reported during this interval. In a second report (Yang et al., 2005), only the first of 4 throat swabs was positive for iVDPV1. The three negative swabs were collected after the patient had also stopped excreting iVDPV1 in stools. Thus, our report is the first report of continued isolation of vaccine-derived poliovirus from oropharyngeal mucosa over an extended period of time. Importantly, it is also the first case of persistent oropharyngeal PV2 infection in PIDs after eradication of wild type 2 poliovirus was declared.

The prevalence of persistent poliovirus infections of oropharyngeal mucosa in PIDs is difficult to determine at present because isolation of poliovirus from oropharyngeal mucosa is underperformed and under-reported. Lack of reporting does not mean that isolation was not tested. Reporting of negative findings for poliovirus isolation from oropharyngeal mucosa in PIDs is important for containment and eradication to indicate that testing was performed and no poliovirus was found.

The case reported here illustrates why hospitalization guidelines for PIDs should include testing of oropharyngeal mucosa. It also illustrates that droplet isolation should be added to recommended initial contact precautions for PIDs when poliovirus is identified in stools until the need is ruled out. Persistent poliovirus infections in PIDs are different from self-limiting infections in immune competent individuals. Therefore guidelines should also take into consideration the frequency of retesting of PID patients and close contacts when a case of poliovirus persistence is identified. Even though risk of transmission of iVDPVs is low, it is still possible (Aghamohammadi et al., 2017; Avellon et al., 2008). It is relatively easy to reduce risk of transmission in hospitals by implementing strict containment protocols. However, it is much more difficult to do so in a community setting for poliovirus-positive PID patients where compliance is voluntary, as in the current case.

Funding

This work was supported by routine operating funds supplied to the Israel National Center for Polio and Non-polio Enteroviruses by the Public Health Service of the Israel Ministry of Health.

Ethics statement

The ERB of Sheba Medical Center approved this study (SMC-3804-16). Written informed parental consent was obtained.

Conflict of interest

The authors declare that the work was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Author contributions

MW, LMS DS and GR conceived and designed the study: JA, LW, IS and IB performed cell culture and molecular diagnosis; MW, LMS, GR, DS, VI and EM contributed to analysis and interpretation of data. RS and TS collected and wrote the clinical case data. MW, LMS and DS drafted the article. All authors approved the final version of the article.

References

Aghamohammadi A, Abolhassani H, Kutukculer N, Wassilak SG, Pallansch MA, Kluglein S, et al. Patients with primary immunodeficiencies are a reservoir of poliovirus and a risk to polio eradication. Front Immunol 2017;8:685.

Alexander JP, Ehresmann K, Seward J, Wax G, Harriman K, Fuller S, et al. Transmission of imported vaccine-derived poliovirus in an undervaccinated community in Minnesota. J Infect Dis 2009;199:391–7.

Alexander Jr. JP, Gary [79_TD$DIFF]Jr HE, Pallansch MA. Duration of poliovirus excretion and its implications for acute flaccid paralysis surveillance: a review of the literature. J Infect Dis 1997;175(Suppl. 1):S176–82.

Avellon A, Cabrerizo M, de Miguel T, Perez-Brena P, Tenorio A, Perez JL, et al. Paralysis case and contact spread of recombinant vaccine-derived poliovirus, Spain. Emerg Infect Dis 2008;14:1807–9.

Bellmunt A, May G, Zell R, Pring-Akerblom P, Verhagen W, Heim A. Evolution of poliovirus type I during 5.5 years of prolonged enteral replication in an immunodeficient patient. Virology 1999;265:178–84.

GPEI. Global eradication of wild poliovirus type 2 declared On-line at http://www.polioeradication.org/mediaroom/newsstories/Global-eradication-of-wild-poliovirus-type-2-declared/tabid/526/news/1289/Default.aspx. [Last accessed on September 2016]. 2015.

Hanna S, Etzioni A. 'MHC class I and II deficiencies. J Allergy Clin Immunol 2014;134:269–75.
Jenkins HE, Aylward RB, Gasasira A, Donnelly CA, Mwanza M, Corander J, et al. Implications of a circulating vaccine-derived poliovirus in Nigeria. N Engl J Med 2010;362:2360–9.
Jorba J, Diop OM, Iber J, Henderson E, Zhao K, Sutter RW, et al. Update on vaccine-derived polioviruses - worldwide, January 2017–June 2018. MMWR Morb Mortal Wkly Rep 2018;67:1189–94.
Khetsuriani N, Prevots DR, Quick L, Elder ME, Pallansch M, Kew O, et al. Persistence of vaccine-derived polioviruses among immunodeficient persons with vaccine-associated paralytic poliomyelitis. J Infect Dis 2003;188:1845–52.
Li L, Ivanova O, Driss N, Tiongco-Recto M, da Silva R, Shahmahmoodi S, et al. Poliovirus excretion among persons with primary immune deficiency disorders: summary of a seven-country study series. J Infect Dis 2014;210 (Suppl. 1):S368–72.
Macklin G, Liao Y, Takane M, Dooling K, Gilmour S, Mach O, et al. Prolonged excretion of poliovirus among individuals with primary immunodeficiency disorder: an analysis of the World Health Organization registry. Front Immunol 2017;8:1103.
Shulman LM, Gavrilin E, Jorba J, Martin J, Burns CC, Manor Y, et al. Molecular epidemiology of silent introduction and sustained transmission of wild poliovirus type 1, Israel, 2013. Euro Surveill 2014;19(7). . pii=20709. Available online: http://www.eurosurveillance.org/ViewArticle.aspx?ArticleId=20709.
Weil M, Shulman LM, Heiman S, Stauber T, Alfandari J, Weiss L, et al. Prolonged excretion of type-2 poliovirus from a primary immune deficient patient during the transition to a type-2 poliovirus-free world, Israel, 2016. Euro Surveill 2016;21.
WHO. Polio Laboratory Manual. 4th edition WHO; 2004 2004. WHO/IVB/04.10.
WHO. World Health Assembly resolution: poliomyelitis. Geneva, Switzerland: World Health Organization. Sixty-Eighth World Health Assembly 2015; Resolution no. WHA 68.3. Available at http://apps.who.int/gb/ebwha/pdf_files/WHA68/A68_R3-en.pdf. [Last Accessed September 2016]. WHO.
Yang CF, Chen HY, Jorba J, Sun HC, Yang SJ, Lee HC, et al. Intratypic recombination among lineages of type 1 vaccine-derived poliovirus emerging during chronic infection of an immunodeficient patient. J Virol 2005;79:12623–34.