Prevalence of Prolonged and Chronic Poliovirus Excretion among Persons with Primary Immune Deficiency Disorders in the Philippines

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ABSTRACT

Objectives. As part of the global initiative to eradicate poliovirus infections this study aims to: (1) estimate the prevalence of vaccine-derived poliovirus excretion among persons diagnosed with primary immune (B-cell or combined B/T-cell) deficiency disorders (PIDD) in the Philippines; (2) describe clinical features of these PIDD patients excreting poliovirus; (3) genetically characterize vaccine-derived polioviruses isolated from persons with PIDDs; and (4) determine the duration of poliovirus excretion among subjects who tested positive for vaccine-derived poliovirus excretion.

Methods. Seventy-one (71) Filipino patients (ages 0-35 years of age) with PIDD were recruited retrospectively and prospectively over a period of 16 months. The study participants, after informed consent and administration of a questionnaire for baseline data, underwent further testing of quantitative immunoglobulin levels (IgG, IgA, and IgM) and stool poliovirus isolation using two stool samples. Stool specimens which tested positive for the poliovirus were sent to the Regional Reference Laboratory in Australia for further characterization by Intratypic Differentiation (ITD) and Vaccine-derived polioviruses (VDPV) real-time PCR. These participants were then monitored on a monthly basis until laboratory tests identified two sequential months of negative poliovirus stool specimens.

Results. Seventy-one (71) patients underwent interview and quantitative serum immunoglobulin testing. However, one patient expired prior to stool isolate collection. This study, then, documented that none of the remaining 70 Filipino individuals (0-35 years old) with confirmed or suspected PIDDs chronically excreted immunodeficiency–associated vaccine-derived poliovirus (IVDPV). One patient who was a recent OPV-recipient excreted poliovirus Sabin-like 1 transiently (less than 1 month) and two patients excreted non polio-enteroviruses.

Conclusions. Chronic and prolonged poliovirus excretion appears to be uncommon among Filipino patients with diagnosed Primary Immunodeficiency Disease Disorders. However, as part of the continuing global initiative for poliovirus eradication, vigilance is still necessary in patients with primary immunodeficiency diseases. Adequate identification of these patients followed by monitoring their capacity for viral excretion and environmental contamination may be necessary to achieve this goal.

Key Words: Oral poliovirus vaccine (OPV); Primary immune deficiency disorders (PIDD); Vaccine-derived polioviruses (VDPV); Immunodeficiency-associated vaccine-derived polioviruses (iVDPV); Wild Poliovirus (WPV)

ABBREVIATIONS

OPV - oral poliovirus vaccine; **PIDD** - primary immune deficiency disorders; **VAPP** - vaccine-associated paralytic poliomyelitis; **VDPVs** - vaccine-derived polioviruses; **iVDPVs** - immunodeficiency-associated vaccine-derived polioviruses; **cVDPV** - circulating vaccine-derived polioviruses; **WPV -** Wild Poliovirus; **AFP** - acute flaccid paralysis; **VPDS-** Vaccine Preventable Disease Surveillance; **SIAs** - supplementary immunization activities; **QIG** - quantitative immune globulins; **CVID** - common variable immunodeficiency disorder; **WHO** - World Health Organization; **DOH** - Department of Health; **RITM -** Research Institute of Tropical Medicine; **UP-PGH** - University of the Philippines - Philippine General Hospital

Introduction

Poliomyelitis is a highly contagious disease that remains a major global health concern. The poliovirus belongs to the enterovirus subgroup of the picornaviridae family. In most infected persons, wild poliovirus (WPV) transiently inhabits the gastrointestinal tract of human hosts and has 3 serotypes (types 1,2,3), with some differences in virulence. It exhibits increased survival in tropical regions with poor sanitation, high population densities and increased incidence of enteric infections.[1] Most polio virus infections (95%) are asymptomatic. However, a few infected individuals (1-2%) develop symptoms of acute flaccid paralysis. Improvements in hygienic practices over time has led to postponement of the development of natural immunity and increased the

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number of susceptibles culminating in the occurrence of epidemics.[2]

In 1988, the World Health Assembly resolved to eradicate poliomyelitis globally. To achieve this target, the Global Polio Eradication Initiative (GPEI) was launched. The underlying strategies of the GPEI are: 1) to improve routine vaccination coverage; 2) to carry out massive immunization campaigns (usually National Immunization Days (NIDs)) to decrease the incidence of poliomyelitis rapidly; 3) to establish sensitive surveillance for acute flaccid paralysis (AFP) to detect all cases of poliomyelitis; and 4) to conduct "mopping-up" immunization activities to eliminate the last reservoirs of poliovirus circulation.[3] In 1988, over 125 countries were considered endemic for the disease with an estimated 350,000 children developing paralytic poliomyelitis.[4] The Philippines was considered, then, to be among the countries endemic for poliovirus with an incidence rate of poliomyelitis cases noted at 1.8 per 100,000 population.[5]

The introduction of Salk's inactivated poliovirus vaccine (IPV) in 1955 decreased the incidence of poliomyelitis cases by approximately 20-fold in the ensuing next 5 years. However, in 1962, live-attenuated trivalent oral polio vaccine (OPV/Sabin) was advocated over IPV since it more closely imitates the effect of natural infections in the induction of type-specific humoral and mucosal immunity. It was then proposed that the oral vaccine would be potentially more effective in interrupting disease transmission. Furthermore, OPV demonstrated advantages in cost, ease of administration and high vaccine efficacy, especially for inducing mucosal immunity, with low number of doses.[6,7,8]

Due to the intensive eradication drive, a 99% reduction in polio cases has been noted since 1988. In the Philippines, the Expanded Programme on Immunization (EPI) was launched in 1976, supplemented by applying the polio eradication strategies starting in 1992. This caused a drastic reduction in reported poliomyelitis cases such that the country was declared to be wild polio virus-free in 2000.[1,3,4,9] This trend was noted to occur worldwide. However, a leveling out in progress of eradications was noted in 2005 with the persistence of polio-virus endemic areas in Nigeria, India, Pakistan and Afghanistan. Reasons for this persistence have been attributed to insufficient political support, suboptimal health systems, operational issues, and most importantly, insurgency and civil war that restricted vaccine access.[1,3,10]

On the other hand, the massive use of OPV may also have contributed to the difficulty in total polio disease eradication around the world for two reasons. First, would be the development of vaccine-associated paralytic poliomyelitis (VAPP) among vaccinees and their close contacts. And secondly, would be the emergence of vaccine-derived polio viruses (VDPV) which have the potential to revert genetically towards wild-type transmissibility and neurovirulence.[8,11]

VAPP is defined as poliomyelitis occurring in a vaccinee between 7 to 30 days after a dose or in contacts 7 to 60 days after exposure. This condition is a rare consequence of oral poliovirus vaccination. However, with global surveillance of WPV infection, VAPP is now more frequently reported than the former. Polio virus-type susceptibility seems to be dependent on immunologic status: immunocompetent patients are at highest risk for type 3 virus VAPP, immunodeficient patients and healthy contacts for type 2 VAPP.[1,6,7,11,12]

In the gut of immunocompetent individuals, Sabin strains replicate for a limited period of time and are excreted usually for periods up to 30 to 40 days. Containing an RNA virus, live attenuated OPV viruses do mutate, and are prone to intratypic and intertypic recombination, potentially leading to the emergence of genetically divergent strains called VDPVs.[6] VDPVs resemble WPVs biologically and have the potential to cause disease. The degree of genetic change in VDPV is dependent upon the length of time of viral replication in the gut of infected carriers whether healthy or not. Most poliovirus genomes differ from the corresponding parental OPV strain by > 1 % nucleotide positions and are estimated to have replicated for at least 1 year after vaccine administration, longer than the normal period of vaccine replication of 4 to 6 weeks. Based on this mechanism, poliovirus isolates are also classified into three categories based on the extent of the nucleotide sequence divergence from the corresponding parental Sabin OPV strain: 1) OPV-like viruses or (<1% divergence for type 1 and 3 and <0.6% for type 2); 2) VDPVs (1-15 % divergent); and 3) WPVs (> 15% divergent).[8,13]

VDPV outbreaks may be influenced by virus transmissibility, population immunity and other socio-economic and environmental conditions. Thus, VDPVs have been further classified into three categories: cVDPV (circulating VDPV), iDVPV (immunodeficient VDPV) and aVDPV (ambiguous VDPV). cVDPVs are transmitted through circulation of OPV-derived virus strains in human populations with deficient herd immunity or low community vaccine coverage. This type of poliovirus is responsible for some re-infections of poliovirus infections in areas previously certified as polio-free. In the Philippines, four new cases of cVDPVs have been identified in 2001, three of which developed poliomyelitis and one was a healthy carrier. These were contained by massive supplemental immunization activities (SIAs) to sustain high immunity in the population. Since then, no reported case has been detected by the Philippine Vaccine Preventable Disease surveillance (VDPS) team.[1,6,8,9,11,13] aVDPV are isolated from paralyzed patients with no evidence of spread to close contacts or are detected from sewage systems.[13]

However, another source of VDPV infection has apparently been traced to patients with primary immunodeficiency diseases (PIDD). Due to poor humoral and mucosal immunity, PIDD patients have been known to harbor poliovirus in their intestinal tracts for prolonged periods leading to chronic excretion of unique, genetically divergent species. This can either happen when an immunodeficient patient receives OPV or through close contact of the immunodeficient patient with a vaccinee. Unlike cVDPVs, iVDPVs may occur in communities with high rates of OPV coverage. Since the introduction of OPV in the 1960s, an increasing number of PID patients have been found to excrete iVDPVs mostly detected only after the onset of AFP.[8] The only way to prevent iVDPV infections is to stop OPV use since there are still no effective anti-viral drugs available that can clear these infections. However, cessation of OPV use may only prevent new cases from occurring if existing iVDPV cases stop excreting spontaneously. Finally, cessation of OPV use could lead to reemergence of cVDPVs in endemic areas and areas of low immunization coverage. Unimmunized individuals in these conditions will then be susceptible to poliovirus infection and, possibly, disease caused by cVDPVs produced by vaccines.[1,6,11,13,14]

PIDD patients whether symptomatic or asymptomatic for poliovirus infections, may therefore serve as potential sources of poliovirus after eradication. Although rare, iVDPV infections could pose as obstacles to global poliovirus eradication since present strategies do not offer any effective solutions to this condition. Cases of prolonged excretion of VDPV by immunodeficient patients in other countries have been well documented based on regular monitoring by WHO.[6,8,15] During the period of July 2009 to March 2011, four patients have been documented to have iVDPV in the Asia-Pacific Region alone.[8] In the Philippines, there has been no study documenting chronic excretion of VDPV in patients with either diagnosed or suspected immunodeficiency.

This study attempted to determine the prevalence of chronic iVDPV excretion among Filipino individuals diagnosed with PIDD and provide information on whether these persons could serve as potential reservoirs of VDPVs in the Philippines.

Study Objectives

1) To estimate the prevalence of vaccine-derived poliovirus excretion among persons diagnosed with primary immune (B-cell or combined B/T-cell) deficiency disorders (PIDD) in the Philippines;
2) To describe clinical features of these PIDD patients excreting poliovirus;
3) To genetically characterize vaccine-derived polioviruses isolated from persons with PIDDs; and
4) To determine the duration of poliovirus excretion among subjects who tested positive for vaccine-derived poliovirus excretion

Methods

Study Population

Clinical Recruitment of Patients

Prospective and retrospective approaches were utilized for the clinic-based recruitment of study participants. Eligible participants were recruited from February 2010 until July 2011.

Prospective recruitment was coordinated through the University of the Philippines - Philippine General Hospital (UP-PGH) Section of Allergy and Immunology in Manila. Patients residing within Metro Manila and some areas of the National Capital Region (NCR) during the study period were referred to UP-PGH for recruitment. Recruitment of PIDD patients residing outside of the greater Manila metropolitan area was coordinated by the study investigators, local immunologists and clinicians.

Retrospective recruitment was from the PIDD registry UP-PGH Section of Allergy and Immunology. Contact information of the patients was gathered from the retrieved medical charts.

Eligible participants for this study were those who met the clinical case definition of PIDD and who were less than 35 years of age. While most persons with PIDDs are diagnosed during infancy and early childhood, common variable immunodeficiency disorder (CVID) may not be discovered until the second decade of life.[16] The 35-year-old age limit was set to ensure that these patients with CVID were appropriately included since diagnosis of this condition occurs in the second or third decade of life. CVID patients, therefore, may have already been given OPV inadvertently early in life when the condition was not yet diagnosed. PIDDs are unlikely to be diagnosed after this age. A physician-suspicion of PIDD was required for eligibility and enrolment.

Definitions

Two definitions of primary immunodeficiency disorders (PIDD) were used in this study.

1) Confirmed Primary Immunodeficiency Disorders (PIDD):
   These include disorders of immunity, affecting B and T-cells that are not associated with other illnesses that impair the immune system.[16] Because PIDD diagnostic criteria may vary,[17] a confirmed diagnosis, for the purpose of this study, were based on clinical assessment, Quantitative Immunoglobulin (QIG) values, other confirmatory

immunologic tests (e.g. T and B cell enumeration) and response to therapy as outlined in the chapter on primary immunodeficiency disorder diagnoses in the Textbook of Pediatrics and Child Health, 4th edition.[18]

2) Probable Primary Immunodeficiency Disorders (PPID):
These included disorders which had increased susceptibility to infections and at least one of the 10 warning signs for primary immunodeficiency presented by the Jeffrey Modell Foundation (Appendix A).[19]

Vaccine Preventable Disease Surveillance Subjects

Patients with positive stool samples from the Philippine Vaccine Preventable Disease surveillance project were included in the study. The 2001 cVDPV outbreak in the Philippines which resulted in three cases of paralytic poliomyelitis was detected through the country's vaccine preventable disease surveillance (VDPS) system. Two stool specimens were supposed to be collected from all AFP cases and examined for polioviruses. Since 2005, all poliovirus isolates underwent standard ITD and the majority had been sequenced to assess whether the detected virus was wild or vaccine-related. Although no isolated Sabin-like viruses have exhibited 1% or more divergence from the original OPV strain (since the 2001 outbreak), recent review of the AFP database has identified four AFP cases whose stool samples tested positive for poliovirus. However, only three had a divergence of >0.5. These viruses may reflect minimal community circulation or prolonged viral replication in a single person, which can occur in individuals with PIDDs.

Hospital patients determined to have transient or secondary immunodeficiency and possible acute flaccid paralysis (AFP) cases no longer residing in the Philippines were excluded from the study. No other exclusion criteria were applied.

Ethical Considerations

A written informed consent (in English or the local dialect Tagalog) was obtained from each study participant or from a legal guardian in case the participant was a minor. For the purpose of this study, any participant less than 18 years of age was considered a minor. An additional assent form (in English or in Tagalog) was obtained from patients 7 to less than 18 years of age. Research assistants were required to sign a document for confidentiality of all study outcomes.

Confirmed and probable PIDD patients were provided treatment and medical counseling after recruitment based on best health care practices by trained experts in the field of allergy and immunology in different health care centers in the Philippines. All patients were provided education regarding his/her PIDD diagnosis including preventive measures for infectious diseases (e.g. avoid live vaccines, hygiene, safe water, good nutrition), genetic counseling for the family, as well as information regarding various treatment options. Participants were allowed to withdraw from the study at any time without negative consequence to continued immune deficiency treatment and/or access to primary health care.

Ethical clearance was sought from the Ethical Review Boards at the Philippine General Hospital in Manila, and the Research Ethics Review Committee (ERC) at WHO Headquarters in Geneva, Switzerland prior to initiation of the study.

Data Collection

Data collection was planned in two phases: 1) initial screening of all participants, and 2) follow-up of participants with detected poliovirus excretion.

1) Initial screening

Following consent, the research assistant or the study investigators administered a standardized questionnaire and collected a single blood sample (2 ml) from each participant. These blood specimens were sent to the UP-PGH laboratory for QIG testing. QIGs were used to assess participants' immunologic status by providing serum measurements of IgG, IgA, and IgM. QIGs were assessed for all patients during the initial screening (see QIG reference ranges in Appendix B). PIDD patients receiving IVIG treatment within the previous three weeks were recruited at initial contact. However, QIG testing was delayed until the next scheduled treatment visit (blood sample collected prior to treatment).

Participants recruited were also provided with a WHO recommended stool collection kit and were asked to collect two separate specimens (8-10g each) at least 24 hours apart. The study followed the WHO protocols for maintaining the reverse cold chain during storage and transport of collected stool specimens. The stool samples were processed at the National Polio Laboratory operated by the Philippines Department of Health at the Research Institute for Tropical Medicine (RITM) using WHO standard guidelines for poliovirus isolation. Poliovirus and the non polio-enteroviruses were isolated in RD (human rhabdomyosarcoma cell line; ATCC CCL 136) and L20B (mouse L). Specimens which tested positive for the poliovirus were sent to the Regional Reference Laboratory in Australia for further characterization by Intratypic Differentiation (ITD) and VDPV real-time PCR. Laboratory data were recorded in a separate form for encoding into the database.

Attending physicians, study participants and their guardians were provided with laboratory test results and educated regarding the interpretation of the findings.

2) Follow-up

The participants who tested positive for the poliovirus were monitored on a monthly basis until laboratory tests identified two sequential months of negative poliovirus stool specimens (for a total of 4 negative samples). An additional questionnaire was administered at the first follow-up contact in order to collect further information on the participant's current medical status and treatment. Two stool specimens were collected using the same protocol as described above starting one-month following availability of results from the initial specimen collection.

Prolonged or long-term (chronic) vaccine poliovirus excretion was defined as either 1) the detection of vaccine-related poliovirus with genetic evidence suggesting at least six-months of replication (sequencing data indicating >0 .5% divergence from original Sabin strain); or 2) the repeated detection of vaccine-related poliovirus from stool specimens collected at least six months or more apart (the same patient has been excreting poliovirus over a six-month period). A vaccine-derived poliovirus was defined as vaccine-related poliovirus isolates with at least 1% nucleotide divergence in the VP1 region from the corresponding vaccine strain, suggesting, on average, one year of replication.[13]

Patients identified to be chronic excreters were to be followed up by the Philippine DOH, AFP surveillance team and local immunologists. They were also given continued health care support based on best health care practices in the Philippines.

Data Management

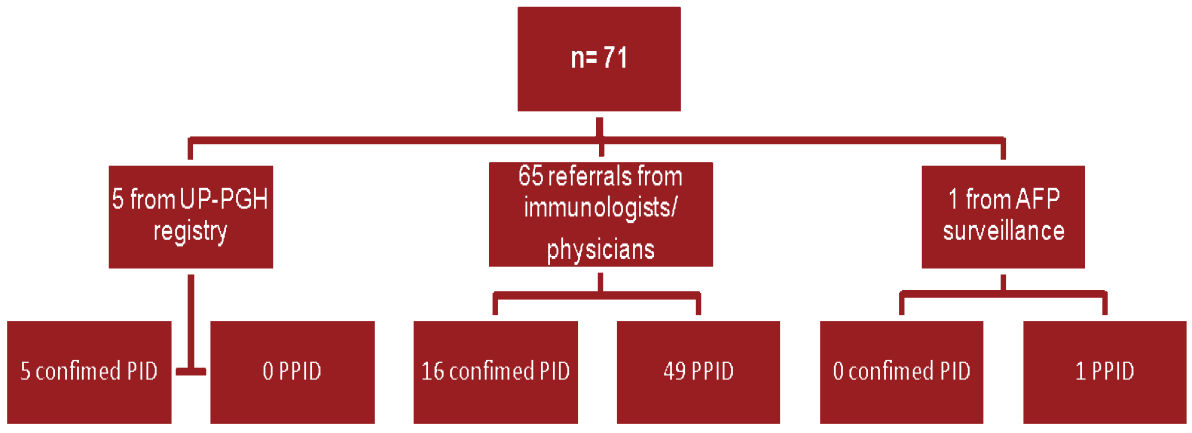
Questionnaire data were double-entered into a Microsoft OfficeAccess database and EpiInfo at UP-PGH. Stool specimen results were forwarded from the National Polio Laboratory to UP-PGH and merged with questionnaire data based on the unique study identifier assigned at the time of enrolment. Univariate statistics were generated in EpiInfo to verify accuracy of data entry. Missing data and/or inconsistencies were identified and corrected. To assure confidentiality, electronic databases and all laboratory results were referenced solely to the unique study ID provided to each participant at the time of the enrolment. The only link between study data and subject identity was the hard copy of the questionnaire. Therefore, original questionnaires are maintained at UP-PGH and copies of all study documentation (electronic and hard copies) will be sent only to the WHO for long-term storage.

Results

A total of 71 patients were recruited for the study. There were five participants from the UP-PGH registry, one participant from the AFP surveillance and 65 participants were referred by immunologists/physicians (Figure 1). There were 38 primary immunodeficiency cases included in the Immunodeficiency registry of the Section of Allergy and Immunology - Philippine General Hospital; however, only 22 charts were available. Five patients from the registry with confirmed PID were located and included in the study. Two patients already died and 15 others have moved residences and were not located.

In the VPDS surveillance, three out of the four patients had poliovirus isolate with a greater than 0.5% divergence. Out of these 3 patients, one had died a year before the start of the study, another had refused further tests even at the DOH surveillance level. Only one of these three patients was included in the study.

Figure 1. Source of Recruitment of Confimed (PIDD) and Probable Primary Immunodeficiencies (PPID)



UP-PGH –University of the Philippines
AFP Surveillance– Acute Flaccid Paralysis Surveillance
PIDD- primary immunodeficiency disorder
PPID – probable primary immunodeficiency

Table 1. Baseline Characteristics of Participants with Primary Immunodeficiency

	Number of Participants	Median Age at Enrolment (years) (range)	Gender		Polio Vaccination Status		
			Male	Female	OPV	IPV	None
Confirmed PID	**21**	**10 (0.33-35)**	**15**	**6**	**16**	**2**	**3**
Common Variable Immunodeficiency	*6*		*4*	*2*	*6*	*0*	*0*
Agammaglobulinemia	*2*		*2*	*0*	*2*	*0*	*0*
Hypogammaglobulinemia	*1*		*1*	*0*	*1*	*0*	*0*
B cell Deficiency	*2*		*2*	*0*	*1*	*1*	*0*
Severe Combined Immunodeficiency	*1*		*1*	*0*	*1*	*0*	*0*
Combined T and B cell Defects	*1*		*1*	*0*	*1*	*0*	*0*
Di George Syndrome	*2*		*2*	*0*	*1*	*0*	*1*
Hyper IgE Syndrome	*2*		*1*	*1*	*1*	*1*	*0*
Chronic Mucocutaneous Candidiasis	*2*		*0*	*2*	*2*	*0*	*0*
Probable Primary Immunodeficiency	***50***	***2 (0.06-16)***	***29***	***21***	***28***	***15***	***7***
Total	**71**		**44**	**27**	**44**	**17**	**10**

Baseline demographic and clinical data were obtained from the 71 participants in the study (Table 1). The median age at time of recruitment was 3 years and range was 23 days to 35 years old. There were more male participants (62%). The majority (87.1%) of the participants had received polio vaccine. From the 61 participants who received polio vaccine, 44 (72.1%) had received OPV while 17 (27.9%) had received IPV. In 22 (31.4%) participants, one or more household members less than five years old had received oral polio vaccine one year prior to stool culture.

Majority of the participants were diagnosed to have PPID (70.4%). (Table1)

Three of the participants initially classified as PPID had undergone further immunologic evaluation by their private immunologists and were subsequently diagnosed with Cartilage Hair Hypoplasia (1), B cell deficiency (1) and T cell deficiency (1). None of those classified as PPID at the start of the study showed QIG levels below normal for age. The patient recruited from the vaccine preventable disease surveillance (VDPS) project developed seizures and some temporary neurologic symptoms 17 days after the 3rd dose of OPV. He was diagnosed to have PPID since an immunologist who saw him noted a history of seizures before the paralytic event and the presence of dysmorphic features (low set ears, micrognathia, difficulty swallowing and small tonsils and normal QIG levels). In Di George Syndrome, aside from the presence of dysmorphic features, hypoparathyroidism may occur; this can thus manifest as hypocalcemic seizures. Furthermore, immunologic parameters are not always deranged; thus serum antibody levels may be normal in the partial type of Di George syndrome. However, this diagnosis must be confirmed by further testing.

There were 21 confirmed PID at the start of the study. Thirteen patients had potential B cell-deficiencies: 6 patients with CVID, 2 patients with agammaglobulinemia, 1 patient with hypogammaglobulinemia, 2 patients with B cell-deficiency, 1 patient with severe combined immunodeficiency and 1 patient with combined T and B cell defects (Table1). The rest of patients had non-B cell PIDD. The study did not limit the participants to those with humoral deficiency as the purpose of the study was to identify poliovirus excreters among those with primary immunodeficiencies in general.

Four patients with B-cell deficiencies (two with CVID, one with hypogammaglobulinemia, one with agammaglobulinemia) had low levels of serum IgG during the time of evaluation. Serum IgA and IgM values were normal for all patients tested.

Four out of six of the CVID participants were receiving IVIG therapy. Those with agammaglobulinemia (2/2), hypogammaglobulinemia (1/1), SCID (1/1) and B cell deficiency (1/2) have also received IVIG therapy in the past.

Virus Isolate Results

70 participants submitted two stool specimens each at least 24 hours apart. Stool specimens were not collected prior to the demise of one PPID participant. One participant with PPID had a poliovirus isolate in the stool culture. Intratypic differentiation and real-time PCR result showed Poliovirus 1 Sabin-like isolate. The isolated poliovirus was consistent with a recently acquired vaccine with less than 1.0% divergence from the parent vaccine viruses based on the nucleotide sequence of the VP1 gene. The participant had received the third dose of OPV six days before the submitted stool specimen. Follow-up stool cultures obtained 1 and 2 months after were negative for poliovirus.

No viruses were isolated from the stool sample of sixty seven (95.7%) participants (Table 2). Non polio-enteroviruses were isolated from two participants with PPID. These viruses were not further characterized. The participants and referring immunologists were informed of the results of the stool culture.

Table 2. Virus Isolates from Patients with Primary Immunodeficiency

	Poliovirus 1 Sabin-like detected	Non polio Enteroviruses detected	No virus detected	Total Stool samples
Severe Combined Immunodeficiency	0	0	1	1
Common Variable Immunodeficiency	0	0	6	6
Hypogammaglobulinemia	0	0	1	1
Agammaglobulinemia	0	0	2	2
Probable Primary Immunodeficiency	1	2	46	49
Chronic Mucocutaneous Candidiasis	0	0	2	2
Combined T and B cell Deficiency	0	0	1	1
DiGeorge Syndrome	0	0	2	2
B cell Deficiency	0	0	2	2
Chronic Granulomatous Disease	0	0	2	2
HyperIgE Syndrome	0	0	2	2
Total	**1**	**2**	**67**	**70**

Stool specimens were collected from the VPDS surveillance patient four days after the onset of acute flaccid paralysis in 2008; Poliovirus types 1 and 3 Sabin-like were isolated at that time from the stool of the patient. The patient developed neurologic symptoms 17 days after administration of the third dose of OPV. There were no viruses isolated from the stool of the same participant 5 days and one month after onset of the AFP. The patient's neurologic symptoms, however, did not persist; hence, he was not diagnosed to have vaccine-associated paralytic poliomyelitis (VAPP). The patient was asked to submit two stool samples for this study; there was no virus isolated from the stool specimen 3 years after his last positive stool culture.

Discussion

This study revealed that there was no prolonged or chronic excretion of polioviruses in Filipino patients diagnosed with confirmed or probable primary immunodeficiency disorders. Majority of our patients had received OPV in the past and one third had been exposed to household contacts who had received OPV one year prior to study evaluation. There was only one patient with probable PID who transiently excreted Sabin-like Poliovirus 1 for less than a week after receiving a third dose of OPV. Two patients yielded non polio-enterovirus strains in their stool cultures.

Immunity to enteroviruses, such as poliovirus, is primarily antibody or B cell-mediated. The mechanisms of increased susceptibility of B cell-deficient patients to these viruses are still unclear. Defects in Toll-Like receptor (TLR) 8 and 9 signaling with impairment of pro-inflammatory cytokines (tumor necrosis factor [TNF–α] and interleukin [IL]-6) production in B cells have been proposed to occur. Because of the complexity of interactions in the form of cellular cross-talk within immune system, defects in one component may affect the immunologic functions of other subsystems involved in the immune response.[19,20,21] Theoretically, lack of T cell-mediated B-cell activation can lead to deficient antibody production. There are some data indicating that immunodeficient patients with potential T cell defects or combined T and B cell defects (such as CVID, SCID) may also be predisposed to chronic poliovirus shedding.[8,22] Evidence also exists that lack of B cell function is not solely responsible for chronic poliovirus excretion.[23,24,25] In this study, we explored the possibility of chronic polio virus excretion in other types of PIDD; however, we did not encounter any patient with this condition among the non B-cell immunodeficiency diseases. This confirms that poliovirus susceptibility may indeed be seen predominantly in patients with humoral deficiencies.

Most patients with B cell deficiencies stop shedding poliovirus spontaneously 1 year or more after infections for unknown reasons.[6] Although rare, immunodeficient persons with defects in humoral or mucosal immunity have been detected to chronically carry and excrete these polioviruses and other enteroviruses for even as long as 10 years.[6,15,17,20,22] So far, 50 persons with B-cell immunodeficiencies have been reported to WHO with polio excretion for 6 months or more.[8] None of our patients with diagnosed B-cell defects such as Hypogammaglobulinemia, Agammaglobulinemia, B-cell deficiency, Common Variable Immunodeficiency (CVID) and Severe Combined Immunodeficiency (SCID) excreted poliovirus even among the four patients with low serum IgG levels. This finding is similar to the study done in the United States, Mexico, Brazil and the United Kingdom in 2004, wherein long-term shedding of individuals with primary B cell deficiencies was not identified.[17] The absence of chronic iVDPV in our group of patients may also be explained by the fact that many of our immunodeficient patients in the Philippines may have low survival rates and, hence, were naturally excluded from the study.

Out of 13 B-cell immunodeficient patients in our study, 9 were given Intravenous Immunoglobulin (IVIG) treatment. This treatment is used routinely as replacement therapy in B-cell deficient patients. Most of our B-cell deficient patients received IVIG treatment which can explain the absence of virus shedding in this group. However, we also observed that even without IVIG intervention, untreated B-cell deficient patients in our study did not shed poliovirus. This supports evidence that IVIG therapy may not always be vital in containing chronic poliovirus infections in immunodeficient patients. Although, it may decrease the risk of paralytic complications of iVDPV in these types of patients, current evidence suggests that it does not prevent nor clear persistent enteroviral infections.[16,23,26] In fact, fluctuating antibody levels during IVIG therapy may even be a factor for rapid antigenic evolution of iVDPVs.[24]

iVDPV infections in immunodeficient patients may or may not lead to vaccine-associated paralytic polio (VAPP) despite their prolonged existence in the affected individual's internal and external environment.[24,27] The risk though of VAPP among immunodeficient patients is ~ 3000-fold higher than in immunocompetent persons.[15]

Immunodeficient patients are most often exposed through OPV vaccination. Some unimmunized individuals, however, also develop infection or paralytic polio.[13] Many die from VAPP or from other concomitant infections; however, there was one patient with X-linked agammaglobulinemia reported to have survived paralytic poliomyelitis with minimal complications.[27] Although VAPP has also been noted to occur in immunocompetent individuals, PID patients are still considered as high risk for VAPP and virus transmission to the general population. Unlike cVDPVs, high herd immunity does not protect the community from iVDPV spread because the latter could arise anytime in an immunodeficient host due to weak mucosal and humoral immunity.[1]

The lone acute paralytic patient included in our study from the VPDS Surveillance project in the Philippines was referred for immunologic evaluation since he excreted a vaccine-like Poliovirus with >0.5% divergence. This is considered potentially divergent towards a VDPV strain in the local DOH surveillance system and hence was carefully monitored for further mutations. The patient, based on clinical evaluation, may have a possible DiGeorge Syndrome, a PID which is characterized by dysmorphic features, occasional low T cell counts but basically normal immunoglobulin levels. This patient's QIG levels were normal and he no longer excreted poliovirus 3 years after last positive stool culture for polioviruses. This patient may need further work up for immunodeficiency such as a cardiac evaluation, T and B cell or IgG subclass determinations in order to diagnose him as having DiGeorge syndrome. It appears, though, that his humoral immune system is intact such that he was able to stop shedding the virus eventually. VAPP has also been noted to occur in immunocompetent individuals.[1]

The majority of our immunodeficient patients were diagnosed to have PIDD. The Philippine health care system, especially government hospitals, do not normally have the facilities or funding to confirm the diagnosis of patients with suspected immunodeficiencies presenting as recurrent infections. We used the Jeffrey Modell Foundations Screening list of "10 warning signs of immunodeficiency" to recruit patients into our study due to financial constraints (Appendix A). We, however, realize the limitation of using this tool since it may not be accurate in diagnosing PID. Problems with lack of validation and specificity may in fact lead to over-diagnosis of this condition. The serum immunoglobulin levels via QIG determination helped us document a possible humoral immunodeficiency in these suspected immunodeficient patients. Remarkably, all 50 PPID patients had normal antibody levels. Even IgA deficiency, which is the most common form of antibody deficiency or PID for that matter, was not detected. To strongly confirm PID in these patients, further testing of T cell and B cell counts by flow cytometry or IgG subclass determination may need to be done in the future. What is also interesting is that two PPID patients excreted non polio-enterovirus despite normal QIG levels. This brings back the question of whether the humoral system alone is responsible for clearing enteroviral infections.

iVDPV infections in PIDD patients are rare and sporadic. One explanation for the low prevalence of chronic excretion of poliovirus in immunodeficient patients is the poor survival rate of patients especially in developing countries wherein supportive IVIG treatment is not always available or accessible. However, this same factor may actually reduce the risk in low-income countries, such as the Philippines, of exposure from chronic iVDPV excretors since potential reservoirs have been removed.[17,23]

Despite the perceived low risk of iVDPVs in causing disease to majority of the world's population, continued surveillance is mandatory to adequately quantify the risk these cases may pose to the overall eradication initiative. The efficacy of anti-viral agents such as pleconaril and capsid-binding agents has not yet been established; and currently, a cure for chronic iVDPV excretion is not available. As long as OPV is used, the problems associated with its use will persist. Chronic poliovirus excreters are a potential source of transmission of polioviruses that may revert to wild-type polio virus. Since the use of IPV-only schedule may be prohibitive for developing countries, this may be a problem that will not have a definite solution until OPV use is discontinued after global polio eradication has been achieved.

Aside from monitoring patients presenting with acute flaccid paralysis and sampling sewage for polioviruses in previously declared polio-free areas like the Philippines, surveillance of virus excretion in confirmed or suspected immunodeficient patients should be part of the strategy to contain the global resurgence of poliomyelitis. The rising incidence of iVDPV-associated infections or VAPP may serve as an impetus to develop effective antiviral agents and establish better supportive international and local healthcare management for PIDD patients in low-income countries like the Philippines. Finally, the cost-effective use of IPV on a global level must likewise be considered as a potential solution should other therapeutic and surveillance methods fail in achieving poliovirus eradication.

Conclusion

This study revealed that there were no chronic excreters of vaccine-derived polioviruses (iVDPV) in Filipino patients with PIDD. Although most of our PIDD patients had

received or been exposed to OPV, the risk of developing iVDPV excretion is rare. However, improved diagnosis of Filipino PIDD patients via more specific diagnostic tools and routine VDPV surveillance may be necessary to further assess the risks of chronic iVDPV infections in the general population.

Acknowledgments

We thank the Philippines, National Epidemiology Center, Vaccine preventable disease surveillance (VDPS) Department of Health staff for their extensive surveillance and assistance in locating AFP cases for study inclusion. We also thank the World Health Organization – Western Pacific Region Office (WPRO), for contributing their technical support and Jeanette Rainey (CDC, Atlanta, Georgia) for research protocol development. Poliovirus isolation was facilitated by Hazel Galang and Analisa Bautista (Virology lab, Research Institute of Tropical Medicine). The serum immunoglobulin assays were performed by the Immunology lab (Medical Research lab), University of the Philippines-PGH. We also would like to thank our research assistants John Gervasio, Christina Setubal, Vanessa Fajardo, Aileen Elorde for their excellent technical assistance. The members of the Philippine Society of Allergy, Asthma and Immunology have also been supportive for patient recruitment. Finally, we would like to thank The Section of Allergy and Immunology and the National Institutes of Health, UP-PGH for all the support for this project.

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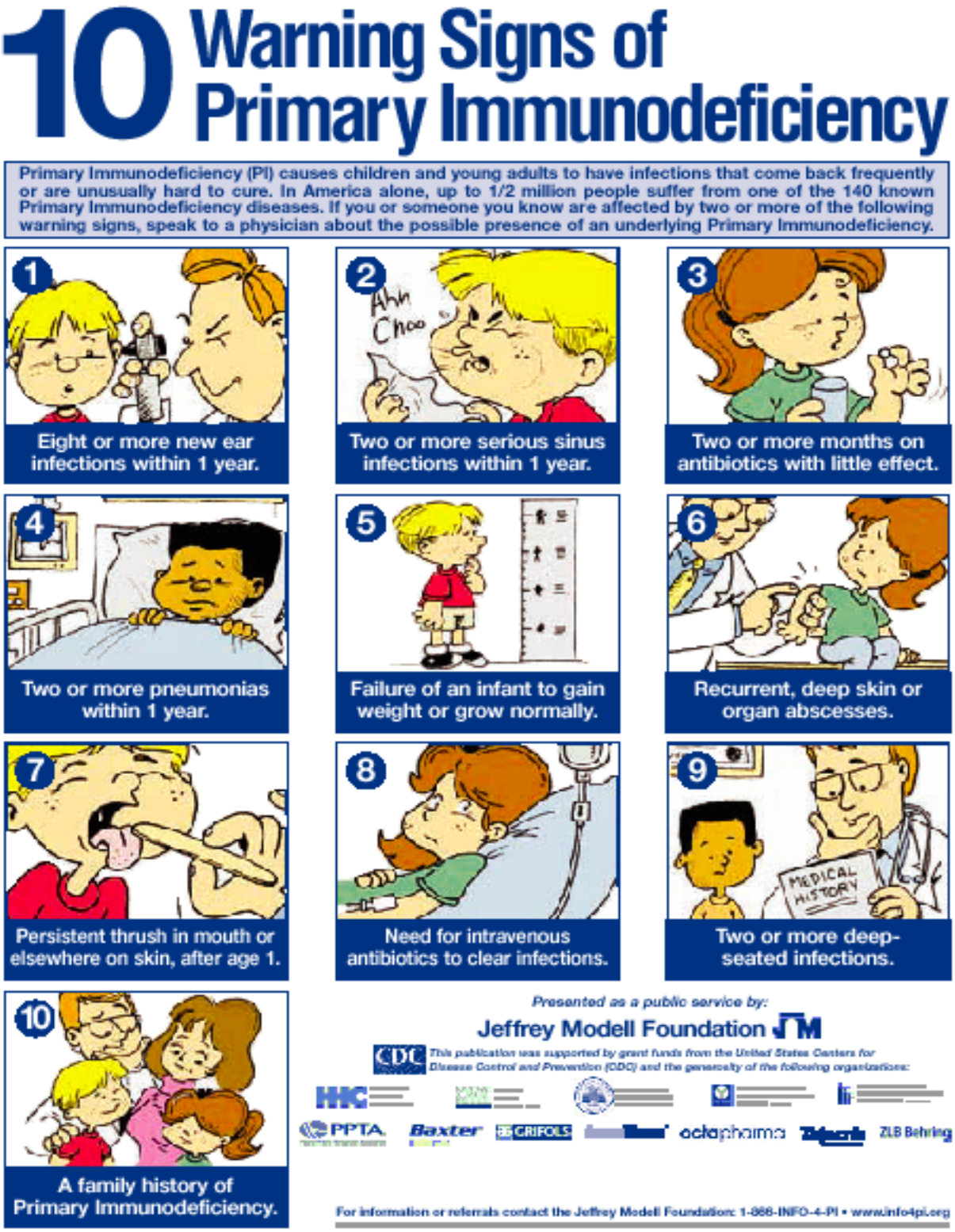
APPENDIX A. Jeffrey Modell Foundation 10 warning signs of Immunodeficiency



APPENDIX B. Serum Immunoglobulin Reference Range

Levels of Immunoglobulins in Sera of Normal Subjects by Age

AGE	IgG mg/dl	IgM mg/dl	IgA mg/dl	Total Ig
Newborn	1031 ± 200	11 ± 5	2 ± 3	1044 ± 201
1-3 mo	430 ± 119	30 ± 11	21 ± 13	481 ± 127
4-6 mo	427 ± 186	43 ± 17	28 ± 18	498 ± 204
7-12 mo	661 ± 219	54 ± 23	37 ± 18	752 ± 242
13-24 mo	762 ± 209	58 ± 23	50 ± 24	870 ± 258
25-36 mo	892 ± 183	61 ± 19	71 ± 37	1024 ± 205
3-5 yr	929 ± 228	56 ± 18	93 ± 27	1078 ± 245
6-8 yr	923 ± 256	65 ± 25	124 ± 45	1112 ± 293
9-11 yr	1124 ± 235	79 ± 33	131 ± 60	1334 ± 254
12-16 yr	926 ± 124	59 ± 20	148 ± 63	1153 ± 69
Adults	1158 ± 305	99 ± 27	200 ± 61	1457 ± 353