Journal of Mucosal Immunology Research

Ahmad et al., J Mucosal Immunol Res 2018, 2:2

Research Open Access

Intestinal Mucosal Immunity against Polioviruses in Adolescent and Adult Populations: A Community Based Cross-sectional Study in India

Mohammad Ahmad[1*], Tapan Dhole[2], Abhishek Kunwar[1], Rakesh Vishwakarma[1], Gopal Krishna Soni[1], Lalitendu Mohanty[3], Raman Sethi[1] and Sunil Bahl[4]

[1]*World Health Organization, Safdarjung Enclave, New Delhi, India*

[2]*Sanjay Gandhi Postgraduate Institute of Medical Sciences, Lucknow, Uttar Pradesh, India*

[3]*Panacea Biotec Limited, Mohan Co-op. Industrial Estate, Mathura Road, New Delhi, India*

[4]*World Health Organization, Regional Office for South-East Asia, World Health House, Indraprastha Estate, New Delhi, India*

[*]**Corresponding author**: Mohammad Ahmad, World Health Organization, Safdarjung Enclave, New Delhi, India. Tel: +91 8800495773; E-mail: ahmadmoh@who.int

Received date: October 23, 2018; **Accepted date:** November 09, 2018; **Published date:** November 15, 2018

Copyright: © 2018 Ahmad M, et al. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

Abstract

Introduction: It is known that the intestinal mucosal immunity against polioviruses wanes with time. Polio free nations risk importation of polioviruses from polio endemic countries and outbreaks of circulating vaccine derived polioviruses. Waning intestinal mucosal immunity against polioviruses in elderly age groups may facilitate setting up of poliovirus transmission, especially in post OPV switch periods. This study assessed the level of intestinal mucosal response among adolescents and adult populations in the high-risk states for poliovirus transmission in India.

Methods: 1200 subjects in two age groups (15-19 years-exposed to multiple doses of OPV and 30-39 years-more exposed to wild polioviruses but almost no OPV) were enrolled in two states of India; Uttar Pradesh and Rajasthan. Subjects were challenged on day-0 and stool samples were subsequently collected on days 7 and 14.

Results: Against expectation, the study found significantly better intestinal mucosal response in the adult (30-39 years) age group compared to adolescents (15-19 years) for poliovirus type 1 and 2. Type-3 showed similar mucosal response in both groups. Residents in Uttar Pradesh, a traditional high-risk state for poliovirus transmission showed better mucosal response compared to those in Rajasthan; significantly so for type-2.

Conclusion: Adult population in high-risk states in India, which was more exposed to wild polioviruses but received almost no OPV doses in past, showed better intestinal mucosal response than the adolescent group which received more OPVs but lesser exposure to wild polioviruses. Despite waning mucosal immunity in the younger population, elderly groups in India have better mucosal immunity, probably induced by infection to wild polioviruses, earlier on, in their life.

Keywords: Intestinal mucosal response; Mucosal immunity; Polioviruses; Vaccine; Gut infection; Sabin virus

Abbreviations: WPV: Wild Poliovirus; RI: Routine Immunization; cVDPVs: circulating Vaccine Derived Polioviruses; IPV: Inactivated Poliovirus Vaccine; tOPV: Trivalent OralPolioVirus; bOPV: Bivalent OralPolioVirus; GPEI: Global Polio Eradication Initiative; SGPGI: Sanjay Gandhi Postgraduate Institute of Medical Sciences; IEC: Institutional Ethics Committee.

Introduction

Despite being the most challenging country for polio eradication and which until 2009 accounted for more than half of the world's polio burden, India successfully eliminated the Wild Poliovirus (WPV) with last reported case in 2011 [1]. Globally, the number of poliomyelitis cases has decreased by >99% since 1988 [2]. Type-2 WPV has been eradicated globally with last detected case in 1999 [3]. No type-3 WPV cases have been detected globally since November 2012 [4]. Type-1 WPV transmission however, continues in Pakistan and Afghanistan [5].

India continues to be at the risk of importing WPV type-1 from polio endemic nations and circulating vaccine derived polioviruses from the countries with recent outbreaks. India also faces the threat of outbreaks of circulating Vaccine Derived Polioviruses (cVDPVs) in areas with low Routine Immunization (RI) coverage [6,7]. To mitigate these risks, India is implementing the global polio endgame strategy-strengthened Routine Immunization (RI), introduced Inactivated Poliovirus Vaccine (IPV) in RI schedule and switched from trivalent OPV (tOPV) to bivalent OPV (bOPV containing only types 1 and 3 Sabin virus) in 2016 [8]. The OPV switch in 2016 carries a heightened risk of emergence of cVDPV type-2 (cVDPV2) due to circulating Sabin virus type-2 from previous vaccination with tOPV and but no immunity against type-2 subsequent to OPV switch as bOPV does not contain Sabin virus type-2 [9].

The Global Polio Eradication Initiative (GPEI) is hard at work to wipe out poliovirus within the next few years [10]. To achieve global polio eradication possible in the near future, all potential channels that may participate in the transmission of poliovirus needs to be plugged. The polio campaigns in India have focused on 0-5 years age group to provide population immunity with OPV [11]. This has provided very good humoral immunity against all three poliovirus types among infants in the high-risk states in India [12]. However, a good intestinal

mucosal immunity against polioviruses is needed to prevent gut infection, subsequent multiplication and excretion and to abort any potential transmission chains in communities. Intensive use of OPV among 0-5 years old can be expected to provide sustained levels of intestinal mucosal immunity in this age group. However, a study in India concluded that the intestinal mucosal immunity appears to wane significantly within a year of vaccination with OPV [13]. Another study in India has shown waning mucosal immunity among children as old as 10 years, who had not received any OPV since last five years compared to younger children still receiving OPVs [14]. From these findings, it may be presumed that the mucosal immunity would have waned still further in age groups beyond 10 years. Waned intestinal mucosal immunity among elderly populations could be a significant threat to the polio endgame strategy. It could facilitate the cross-country exchange of polioviruses, especially through elderly populations lacking mucosal immunity. Available evidence strongly suggests the likelihood of older individuals participating in the international importation of poliovirus through modern travel and migration [15]. Evidences from Brazil, Israel, Namibia and China, suggest that adult populations participate in poliovirus transmission due to relative a lack of intestinal mucosal immunity at this age [16-20].

Though India conducted many studies to assess the humoral immunity in the high-risk areas for polio transmission, no serious attempts seems to have been undertaken to assess the intestinal mucosal immunity among the elderly populations, either in India or globally. Analyzing the excretion, post OPV challenge is the gold standard of assessing the mucosal response. The Presence of Sabin virus in stool samples post OPV challenge indicates a relative lack of mucosal immunity compared to those who don't excrete the Sabin virus [21]. To examine if mucosal immunity wanes further with age, we conducted a cross-sectional assessment of intestinal mucosal immunity against all three poliovirus types in the adolescent age group (15-19 years) and adult population (30-39 years) in the Indian states of Uttar Pradesh (UP) and Rajasthan. At the time of the study, the adult age group (born before 1995 in the pre-polio campaign era) was not exposed to OPV in their childhood either through polio campaigns or poor RI services existing then. However, the adolescent population (born in or after 1995-post polio campaign period) had received multiple doses of OPV through campaigns and better routine services. The study was implemented during September-October, 2015 and laboratory testing completed in early 2017.

Methods

We assessed the intestinal mucosal immunity by analyzing the excretion pattern of all three Sabin virus serotypes after challenge with tOPV. Day-0 baseline stool sample was collected just prior to the tOPV challenge followed by two samples on 7th and 14th day.

Area selection for the study

We selected two states with different categories of risk for poliovirus transmission for the study. Uttar Pradesh (UP) was selected as a traditional high-risk area for poliovirus transmission. Ten districts in UP with highest number of Wild Poliovirus (WPV) cases during 2005-2011 were selected for the study.

Rajasthan was selected as a risk state with poor routine immunization coverage among states with significant population. 10 districts from Rajasthan with lowest Routine Immunization (RI) coverage in the state during 2012-2013 were selected for the study [22]. In the first stage, one block was randomly chosen from all the blocks in the study district, making 10 blocks each in UP and Rajasthan (total 20 blocks). A typical block in Uttar Pradesh and Rajasthan has about 80-100 polio supplementary immunization activity (SIA) polio campaign teams, each of which cover about 300-500 households during the vaccination campaigns. Twelve SIA team areas were randomly selected for study subject selection from all SIA teams in the block. This makes a total of 120 (12 X 10) study clusters each in UP and Rajasthan.

Sample size and subject selection

Based on the expected proportion of subjects excreting the Sabin virus, post tOPV challenge, a sample of 600 subjects (240 in 15-19 years and 360 in 30-39 years age group) was considered in each state (1200 subjects in both states); providing 7% precision at 95% CI. Five subjects were enrolled from each selected SIA team cluster (2 from 15-19 years and 3 from 30-39 years) to attain a sample size of 600 (5 subjects X 12 SIA team cluster X 10 districts) in each state. The inclusion criteria included-individuals in the age groups of 15-19 years and 30-39 years, those who resided in the district block till 5 years since birth, who were available during all study days and provided consent to participate in the study. Exclusion criteria included individuals with history of chronic constipation and sick individuals requiring hospitalization or undergoing treatment for a major illness.

Study processes

Polio vaccination campaign teams are aided by polio micro plans which help them to move house to house in SIA areas (villages/towns) in a predefined sequential manner. These teams do wall marking at each household visited by them. A few days prior to actual study, study staff comprising of WHO India-National Polio Surveillance Project (NPSP) Surveillance Medical Officers and other government staff, visited the study clusters and starting with a randomly provided first household, moved in the same sequence as that of the polio micro-plans. In the process, the field staff screened and listed inhabitants who were age eligible for the study. About twice the number of age eligible persons, as needed to be enrolled in the study, was listed during the phase to account for absence, refusal to participate on the day of actual study. On the day prior to the visit to the health facility (a government facility), the study staff revisited the households with age eligible persons, explained the study to the family and selected healthy participants based on the inclusion/exclusion criteria. Upon agreement to participate in the study, the study staff provided the subjects with logistics for the stool sample collection and explained the process of collecting and storing the sample in proper cold chain. A study staff visited these households again the next morning to collect the voided stool sample and transported the subjects to the health facility for further study procedures. At the study site, a study physician obtained written informed consent and carried out a medical examination, completed a short questionnaire and administered the challenge dose of tOPV. The two follow up stool samples were similarly collected on the 7th and 14th day of the tOPV challenge.

The stool samples collected at homes were kept in the stool carrier with conditioned ice packs to maintain a temperature of 4-8˚C in the field. These samples were transferred to deep freezer to maintain temperature <-20˚C at the district/block headquarters. At the end of the study, these samples were shipped in dry ice to the polio laboratory at Sanjay Gandhi Postgraduate Institute of Medical Sciences (SGPGI),

Lucknow for further testing. All samples were anonymized before being sent to SGPGI with only subject ID labels on the stool containers. SGPGI polio laboratory is a part of the Polio Laboratory Network in India and is accredited by WHO [23]. At SGPGI, the stool samples were examined for the presence or absence of all three types of Sabin polioviruses following standard protocols from the WHO [24].

The study protocol was approved by the Institutional Ethics Committee (IEC) of Sanjay Gandhi Postgraduate Institute of Medical Sciences (SGPGI), Lucknow and the Research Review Committee (RRC) at WHO-South East Asia Regional Office, New Delhi. Confidentiality of the data collected was maintained and any identifying personal information was not included during data entry or analysis.

Results

Enrolment details: The study team screened 9419 households in the states of Rajasthan and Uttar Pradesh to list 2523 age eligible individuals. Of this, we enrolled 1200 individuals who provided stool samples for day-0 visit (240 in 15-19 years and 360 in 30-39 years age group in each state) and challenged them with tOPV. On day-7, 1198 subjects provided the required stool sample, while 1199 subjects provided the day-14 stool sample. A total of 1198 subjects (99.8%) provided all three stool samples for laboratory testing and their results were included for further analysis (Table 1).

Age Group (years)	15-19	30-39	Total
Uttar Pradesh	240	359	599
Rajasthan	240	359	599
Total	480	718	1198

Table 1: Number of subjects attending all study days and providing all stool samples.

Overall, 9.4% children in 15-19 years and 81.8% in the 30-39 years age group did not receive any OPV. Corresponding figures for Rajasthan are 7.5% and 67.4% and for Uttar Pradesh-11.3% and 96.1%. Uttar Pradesh had more (50.0%) participants in 15-19 years receiving 6 or more doses compared to Rajasthan (37.9%), as seen in Table 2.

State	OPV doses receive Age(Years)	0		1-5		6-10		>10		Unknown		Total
		N	%	N	%	N	%	N	%	N	%	
Rajasthan	15-19	18	7.5	106	442	56	23.3	35	14.6	25	10.4	240
	30-39	242	67.4	7	1.9	1	0.3	0	0.0	109	30.4	359
	Total	260	43.4	113	18.9	57	9.5	35	5.8	134	22.4	599
Uttar Pradesh	15-19	27	11.3	58	242	44	18.3	76	31.7	35	14.6	240
	30-39	345	96.1	12	3.3	0	0.0	0	0.0	2	0.6	359
	Total	372	62.1	70	11.7	44	7.3	76	12.7	37	6.2	599
Total	15-19	45	9.4	164	342	100	20.8	111	23.1	60	12.5	480
	30-39	587	81.8	90	2.6	1	0.1	0	0.0	111	15.5	718
	Total	632	52.8	183	15.3	101	8.4	111	9.3	171	14.3	1198

Table 2: Number of OPV doses received by study participants by age groups and state.

At baseline, before any tOPV challenge (day-0), only one subject (16 years of age) from UP was Sabin virus (type 2) positive and subsequently negative for all three serotypes on day 7 and 14. On day-7 after challenge with tOPV, the Sabin virus excretion among all subjects (both age groups) from both states combined was 30.1%, 31.4% and 22.7% for Sabin virus types 1, 2 and 3 respectively. Corresponding figures for day-14 excretion was 9.4%, 2.3% and 10.3%. State wise and age wise proportion of subjects excreting various serotypes of Sabin virus is depicted in Table 3. The Sabin virus excretion for type-1 at day 14 was 14.2% and 6.3% for 15-19 years and 30-39 years age group respectively. Corresponding figures for type-2 were 2.1% and 2.4% and for type-3 6.5% and 12.8%.

Study day	DAY 7 excretion of Sabin poliovirus types after tOPi/ challenge					DAY14 excretion of Sabin poliovirus types after tOPV challenge				
Sabin virus type	Age (years)	Rajasthan	Utter Pradesh	Total	P-value (States)	Age years)	Rajasthan	Utter Pradesh	Total	P-value (States)
Type 1	15-19	106	98	204		15-19	38	30	68	
	30-39	85	71	156		30-39	24	21	45	
	Total	191	169	360		Total	62	51	113	

	15-19 30.39 Total	44.2 23.7 31.9 0.00	40.8 19.8 28.2 0..00	42.5 21.7 30.1 0.00	0.45 0.21 0.16	15-19 30.39 Total	15.8 6.7 10.4 0.00	12.5 5.8 8.5 0.00	14.2 6.3 9.4 0.00	0.31 0.61 0.26
Type 2	15-19	93	77	170		15-19	7	3	10	
	30-39	129	77	206		30.39	9	8	17	
	Total	222	154	376		Total	16	11	27	
	15-19 30.39 Total	38.8 35.9 37.1 0.47	32.1 21.4 25.7 0.00	35.4 28.7 3 1.4 0.01	0.12 0.00 0.00	15-19 30-39 Total	2.9 2.5 2.7 076	1.3 2.2 1.8 0.42	2.1 2.4 2.3 0.73	0.22 0.79 0.29
Type 3	15-19	51	56	107		15-19	15	16	31	
	30-39	97	68	165		30-39	56	36	92	
	Total	148	124	272		Total	71	52	123	
	15-19 30-39 Total	21.3 27.0 24.7 0.11	23.3 189 20.7 0.19	22.3 23.0 22.7 0.77	0.59 0.00 0.09	15-19 30-39 Total	6.3 15.6 11.9 0.00	6.7 10.0 8.7 0.16	6.5 12.8 10.3 0.00	0.85 0.02 0.06

Table 3: State wise and age group wise pattern of excretion of Sabinvirus on study days 7 and 14 after challenge with tOPV.

On day-7, the overall Sabinvirus excretion in both states combined was significantly lower in 30-39 years age group compared to 15-19 years for type-1 (21.7% *vs* 42.5%, p=0.00) and type-2 (28.7% *vs* 35.4%, p=0.00) but similar for type-3 (23.0% *vs* 22.3%, p=0.77) as shown in Figure 1. Uttar Pradesh had less number of elderly subjects excreting Sabin virus compared to younger group for all three serotypes. Rajasthan too shows similar trend for types 1 and 2, but more number of subjects in the adult group excreted Sabin virus compared to the adolescent subjects, though not statistically significant (27.0% *vs* 21.3%, p=0.11) (Figure 2). On day-7, Sabinvirus excretion for both age groups included was lesser in Uttar Pradesh compared to Rajasthan (significantly for type-2) for all three serotypes [type-1 (28.2% *vs* 31.9%, p=0.16), type-2 (25.7% *vs* 37.1%, p=0.00), type-3 (20.7% *vs* 24.7%, p=0.09)]; as observed from Figure 3. Similar trend was observed for day-14 excretion, though not statistically significant for any serotypes type-1 (8.5% *vs* 10.4%, p=0.26), type-2 (1.8% *vs* 2.7%, p=0.29), type-3 (8.7% *vs* 11.9%, p=0.06) (Table 1).

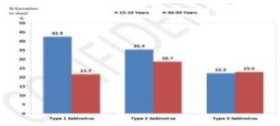

Figure 1: Proportion of Sabinvirus excretors in stool, 7 days after tOPV challenge (both states combined).

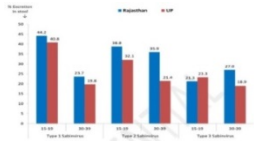

Figure 2: Age and state wise proportion of excretion of Sabinvirus in stool, UP and Rajasthan 7 days after tOPV challenge.

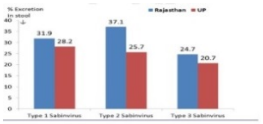

Figure 3: Statewise comaprison of Sabinvirus excretion in stool on day-7 after tOPV challenge.

Discussion

India adopted Expanded Program on Immunization (EPI) since 1978. However, coverage with OPV remained low until better organized national immunization campaigns were initiated in 1995. While Uttar Pradesh conducted multiple polio vaccination campaigns apart from two National campaigns (NID), Rajasthan conducted only two NIDs. Of the reported 5451 polio cases reported in India during 2000-2011, 3.0% occurred in persons >5 years; the highest age being 26 years [25]. Due to relatively lesser polio cases above 5 years, the polio

campaigns in India have focused on a 0-5 years age group to provide population immunity.

Type 2 Wild Polio Virus (WPV2) has been eradicated globally with the last case detected in India, in 1999 [26]. The continued use of tOPV thereafter in the polio eradication program caused paralysis from the vaccine derived polio virus as well as the vaccine associated paralytic poliomyelitis from type-2 Sabin virus (OPV2).

Globally, Sabin type 2 components of tOPV is responsible for the vast majority of circulating VDPV (cVDPV) outbreaks accounting for >95% of all cVDPV cases in the last few years, and approximately 40% of Vaccine-Associated Paralytic Poliomyelitis (VAPP) cases [27].

Stopping the use of tOPV and instead using the bOPV, besides eliminating the risk of type 2 VDPV and VAPP, will provide an additional push to eradicate wild poliovirus types 1 and 3 by virtue of superiority of bOPV over tOPV [28]. The Global Polio Eradication Initiative (GPEI) set achieving high population immunity for type 2 prior to switch as one of the priorities for risk mitigation against tOPV-bOPV switch [29]. Inactivated Poliovirus Vaccine (IPV) has been introduced in OPV only using countries to compensate for loss of type-2 immunity from OPV2 after the OPV switch. WHO recommended at least 1 dose of IPV in the immunization schedule in these countries.

The primary purpose of the IPV dose is to maintain immunity against type 2 poliovirus during and after the global withdrawal of OPV2. Adding an IPV dose is also expected to boost both humoral and mucosal immunity against poliovirus type 1 and 3 and hasten the eradication of these WPVs [30].

Adult subjects in the study were born before the NIDs and as expected received very few OPV doses. More adolescents (born after SIAs introduced in India) from Uttar Pradesh received OPV doses than Rajasthan; as expected due to a greater number of SIAs in Uttar Pradesh than Rajasthan.

The present study shows, against expectation, that the level of intestinal mucosal immunity against polioviruses has not waned among adult Indian population compared to adolescents. The mucosal immunity against type 1 and 2, is in fact, found to be significantly better among adults compared to adolescents and type 3 mucosal response similar in adolescent and adults.

The level of mucosal immunity is better in Uttar Pradesh compared to Rajasthan for all three serotypes, but significantly so for type-2. Uttar Pradesh has better mucosal immunity compared to Rajasthan in both age groups and for all serotypes except 15-19 years age group for type-3. Type-2 excretion is also lower among adults, especially so in Uttar Pradesh with minimal excretion on day-14.

In the settings of rampant Wild Polio Virus (WPV) transmission in the pre-campaign era (pre-1995), it seems exposure of the elderly cohort to WPV rather than to OPVs has provided better lasting mucosal immunity; much better than multiple doses of OPV taken early on in the life by the younger cohort in the study. Not much information is available on this subject matter, though few sources in the past have indicated better mucosal immunity following natural infections compared to immunization with OPV [31-34].

India initiated Acute Flaccid Paralysis (AFP) surveillance for detection of poliomyelitis cases in 1997 when more than 50% of India's polio cases were reported from Uttar Pradesh [35]. Rajasthan had relatively much lesser number of polio cases compared to Uttar Pradesh. In 2009, about 97% (80% in Uttar Pradesh and 17% in Bihar) of the cases were reported from two states [36].

The extremely high force of transmission (FOT) of WPVs in Uttar Pradesh and Bihar may well have been associated with the highest density of infant population in India [37-39]. This historically more exposure to WPVs may be the reason that adult populations from Uttar Pradesh have better mucosal response compared to those in Rajasthan.

Conclusion

Deficits in type-2 mucosal immunity after tOPV-bOPV switch is one of the major obstacles currently facing the polio endgame.The findings from this study along with the high type-2 humoral immunity in India in the high-risk states in 2016 just prior to the tOPV-Bopv switch is re-assuring against the emergence of cVDPV2 in the immediate aftermath of the switch, especially in the traditional high-risk areas for polio transmission such as Uttar Pradesh and areas with similar epidemiological settings.

However, as a matter of caution it should be noted that, as the present adolescent cohort moves in to adult age, they shall build up a cohort of population with waned mucosal response. Also, humoral immunity for type-2 poliovirus post OPV switch would be dependent on inactivated poliovirus vaccine (IPV) coverage in the routine immunization. Areas with low IPV coverage and increasing cohort of population with reduced mucosal response may increase the susceptibility to any type-2 poliovirus importations from countries with recent outbreaks of cVDPV2 and countries with ongoing WPV type-1.

To mitigate these risks, India should think of vaccinating travelers to and from countries with cVDPV2 outbreak with IPV, along with bOPV that is being already administered before the departure to India of passengers from the polio endemic and re-infected countries.This age consideration should be accounted for in any response/intervention against any polio 'event' in future. Studies are needed to compare the level of intestinal mucosal immunity in countries not exposed historically to WPV as intensely as in India.

Acknowledgement

We thank the Ministry of Health and family Welfare, Government of India and the Governments of Rajasthan and Uttar Pradesh for their administrative, logistical and manpower support in the study. The role of Government functionaries, including the district health authorities and Medical Officers is greatly appreciated. We acknowledge the dedicated involvement of ANMs, AWWs, ASHAs, Health Supervisors and other health functionaries in the study.

We thank the Technical Team at the WHO India, National Polio Surveillance Unit, New Delhi, India for their technical inputs and support. We deeply appreciate and thank the involvement of Surveillance Medical Officers, Regional Team Leaders and Sub-regional Team Leaders of the World Health Organization-National Polio Surveillance Project, who participated in the study and very diligently coordinated the field implementation across multiple districts.

We are also thankful to Panacea Biotec Ltd., New Delhi for providing challenge dose tOPV for the study. Finally, we are grateful to laboratory personnel at Sanjay Gandhi Institute of Post Graduate Institute for testing stool samples.

Disclaimer

The authors from the World Health Organization alone are responsible for the views expressed in this article and they do not necessarily represent the decisions, policy or views of the World Health Organization. The authors alone are responsible for the views expressed in this article and they do not necessarily represent the views, decisions or policies of the institutions with which they are affiliated.

References

1. World Health Organization (2014) Polio-free certification: WHO South-East Asia. 7th Meeting of the South-East Asia Regional Certification Commission for Polio Eradication (SEA-RCCPE). New Delhi, India.
2. World Health Organization (2018) Poliomyelitis. Department of Health Services.
3. Centers for Disease Control and Prevention (2001) Apparent global interruption of wild poliovirus type 2 transmission. MMWR Morb Mortal Wkly Rep 12: 222-224.
4. http://polioeradication.org/news-post/three-years-with-no-wild-poliovirus-type-3/
5. Global Polio Eradication Initiative (2018) Wild poliovirus type 1 and circulating vaccine-derived poliovirus cases. Department of Vaccine.
6. Centers for Disease Control and Prevention (2018) Assessing the Risks for Poliovirus Outbreaks in Polio-Free Countries-Africa, 2012 – 2013. MMWR Morb Mortal Wkly Rep 37: 768-772.
7. Jaume J, Ousmane M, Jane I (2017) Update on Vaccine-Derived Polioviruses-Worldwide, January 2016-June 2017. MMWR Morb Mortal Wkly Rep 43: 1185-1191.
8. http://www.searo.who.int/entity/immunization/topics/polio/polio_india_nid_2013/en/
9. World Health Organization (2018) Polio Eradication and Endgame Strategic Plan 2013-2018. Department of the Global Polio Eradication Initiative.
10. Lien G, Heymann DL (2018) The Problems with Polio: Toward Eradication. Infect Dis Ther 2: 167-174.
11. John JT, Vashishtha VM (2018) Eradicating poliomyelitis: India's journey from hyperendemic to polio-free status. Indian J Med Res 5: 881-894.
12. Ahmad M, Bahl S, Kunwar A (2016) Cross-sectional Serologic Assessment of Immunity to Poliovirus in Differential Risk Areas of India: India Seroprevalence Survey-2014. Indian Pediatr 53: s14-19.
13. Grassly NC, Jafari H, Bahl S (2012) Waning intestinal immunity following vaccination with oral poliovirus vaccines in India. J Infect Dis 205: 1554-1561.
14. Jafari H, Deshpande JM, Sutter RW, Bahl S, Verma H, et al. (2014) Polio eradication. Efficacy of inactivated poliovirus vaccine in India. Science 345: 922-925.
15. World Health Organization (2018) Considerations for vaccination recommendations for travelers from polio infected countries. Department of Immunization.
16. World Health Organization (2018) Emergencies preparedness, response. Detection of poliovirus in sewage, Brazil. Department of Immunization.
17. World Health Organization (2018) Global Alert and Response. Poliovirus detected from environmental samples in Israel-update. Emergencies preparedness, response.
18. Centers for Disease Control and Prevention (2006) Outbreak of polio in adults-Namibia, 2006. Morb Mortal Wkly Rep 44:1198-1201.
19. http://www.wpro.who.int/china/mediacentre/factsheets/polio/en/
20. The Global Polio Eradication (2018) Poliomyelitis in China. Department of Health.
21. Wright PF, Alter WW, Ilyushina NA, Hoen AG, Arita M, et al. (2014) Intestinal Immunity Is a Determinant of Clearance of Poliovirus After Oral Vaccination. J Infect Dis 10:1628-1634
22. The Government of India (2018) Annual Health Survey (AHS) 2012-13 Fact Sheet. New Delhi, India.
23. http://www.searo.who.int/india/poliomyelitis/topic/lab_network/en/
24. World Health Organization (1994) Polio laboratory manual. (4th edition), Geneva, Switzerland.
25. World Health Organization (2014) India-National Polio Surveillance. New Delhi, India.
26. Centers for Disease Control and Prevention (2011) Update on vaccine-derived polioviruses--worldwide, July 2009-March 2011. MMWR Morb Mortal Wkly Rep 25: 846-850.
27. Sutter RW, John TJ, Jain H, Agarkhedkar S, Ramanan PV, et al. (2010) Immunogenicity of bivalent types 1 and 3 oral poliovirus vaccine: a randomised, double-blind, controlled trial. Lancet 376: 1682-1688.
28. The Global Polio Eradication Initiative (2018) OPV Cessation. Plan for tOPV Campaigns Immediately Prior to tOPV to bOPV Switch. Department of Immunization.
29. http://www.who.int/wer
30. Ghendon Y, Robertson SE (1994) Interrupting the transmission of wild polioviruses with vaccines: immunological considerations. Bull World Health Organ 6: 973-983.
31. Herremans TM, Reimerink JHJ, Buisman AM, Kimman TG, Koopmans MPG (1999) Induction of mucosal immunity by inactivated poliovirus vaccine is dependent on previous mucosal contact with live virus. J Immunol 8: 5011 - 5018.
32. Sutter RW, Kew OM, Cochi SL, Plotkin SA, Orenstein WA (2004) Poliovirus vaccine-live, Vaccines, 2004. Saunders Company, Philadelphia USA, pp. 651-705.
33. Hird TR, Grassly NC (2012) Systematic Review of Mucosal Immunity Induced by Oral and Inactivated Poliovirus Vaccines against Virus Shedding following Oral Poliovirus Challenge. PLoS Pathog 4: e1002599.
34. Sarkar S (1998) Progress Toward Poliomyelitis Eradication-India, 1998. MMWR Weekly 37: 778-781.
35. Arora NK, Chaturvedi S, Dasgupta R (2010) Global lessons from India's poliomyelitis elimination campaign. Bulletin WHO 88: 232-234.
36. Wright PF, Connor RI, Alter WFW, Hoen AG, Boesch AW, et al. (2016) Vaccine-induced mucosal immunity to poliovirus: analysis of cohorts from an open-label, randomised controlled trial in Latin American infants. Lancet Infect Dis 16: 1377-1384.
37. Indian Expert Advisory Group (2018) Presentation on 'update on polio research studies in India' polio eradication meeting. New Delhi, India.
38. Ministry of Health and Family Welfare (2014) Requirements of polio vaccination for international travellers between India and polio-infected countries. Department of Vaccine.
39. World Health Organization (2018) Keeping the polio virus at bay. Department of Health.