

The Virology of the Polio Virus

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INTRODUCTION

The poliovirus is the aetiological agent of the disease poliomyelitis- a disease that has been known from time immemorial to paralyse and cripple children as well as adult, rich and poor, literate and illiterate. The disease is characterized by acute flaccid paralysis of any of or rarely both of the limbs. The fatal form may involve the medulla and brain stem where the virus causes bulbar poliomyelitis [1].

The poliovirus had been responsible for many epidemics since the 18th century. Epidemic poliomyelitis as a modern disease occurred in Europe in the mid 1800s and in North America in the 1890s [2].

Since then, there had been some landmarks in the study of the virus. In 1905, Landsteiner and Popper [3] transmitted the virus to monkeys by the intracerebral inoculation of the human brain tissue homogenates. Enders [4] cultured the virus in non-neural tissue, thereby eliminating animals for pathogenic studies. This simple discovery marked the beginning of significant progress in the study of the structure, epidemiology, control and prevention of poliomyelitis. This study eventually led to the discovery of the three serotypes and the successful development of vaccines from the virus [5,6]. The most important development in the fight against the poliovirus and in the history of poliomyelitis was the introduction of polio vaccines by Salk and Sabin [7,8].

In 1981, poliovirus became the first RNA virus genome to be molecularly cloned and sequenced. Recent development has included the cloning and sequencing of several strains of the three types of poliovirus [9,10,11,12]. These techniques

have made it possible to determine the precise viral coat amino acids that induce antibody techniques. The poliovirus came to limelight and became a virus of global health importance when the World Health Assembly (WHA) in 1979, following the successful eradication of small pox, decided to target the virus for eradication [13]. Since then tremendous data, information and knowledge have been accumulated on the biology, structure and molecular structure of the virus.

This review article looks into some of the properties and recent knowledge of the poliovirus.

The Structure and Biology of the Poliovirus

The poliovirus is a sub-microscopic intracellular, obligate, non-enveloped icosahedral- shaped virus of diameter between 27-30nm. It is a human enterovirus belonging to the viral family Picornaviridae. The picornaviruses are the smallest of the RNA viruses.

The virus exists in three well-defined serotypes- types 1,2, and 3 and infects cells via a specific receptor – PVR: CD-155. The receptor is a protein which is part of the immunoglobulin super family and is present on the extracellular and intramembranous regions of cells of human origin. This property accounts for the reason why man is the only host for the virus [14].

The RNA genome of the poliovirus consist of a single messenger active strand which is polyadenylated at the 3' terminal and carries a small protein called Vpg at its 5' untranslated (ntr) region. The RNA encodes a large polyprotein which is cleaved into three precursor proteins-the capsid proteins P1 and two non-capsid proteins- the morphogenic proteins P2 and the protease and replicase P3. These precursor proteins are cleaved into 4,3 and 4 end products [14] (Fig. 1).

The poliovirus contains 4 polypeptides chains:

VP1, VP2, VP3 and VP4. The fifth protein, the

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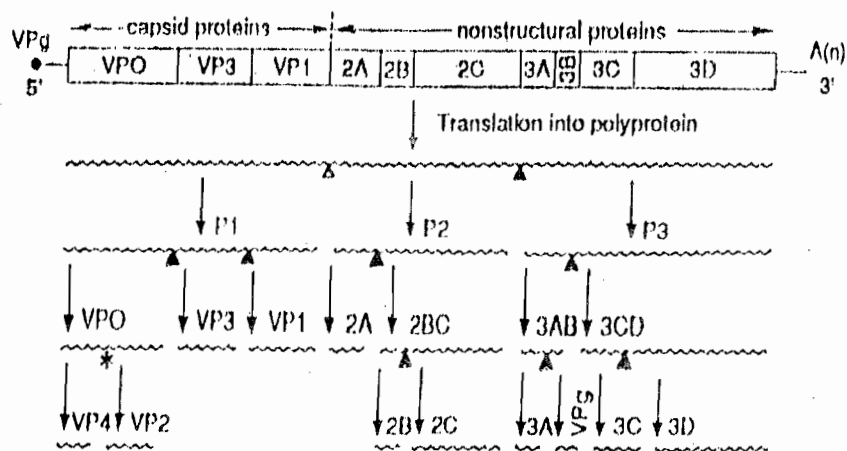


Fig.1: Poliovirus RNA and posttranslational processing of the poliovirus polyprotein

VP0, is the precursor which is cleaved into VP4+VP2 during viral maturation. One of the functions of these viral sub-units is the determination of the host range tissue tropism i.e. disease pathology and antigenicity of the virus [14].

Antigenic Characteristics of Poliovirus

The polio virus consist of three antibody defined types tagged serotypes 1, 2, and 3 [15]. Although, these three serotypes share some antigens, they are however characterized by marked intertypic differences [16].

The viral neutralizing epitopes are located on the three external structural capsid proteins – VP1, VP2, and VP3. Within a serotype, antigenic differences may occur between different isolates [16]. The virus type is actually defined by the capsid encoding sequences that are highly conserved. The non-capsid and non-coding sequences are not highly conserved and may recombine with other enteroviruses in the community. Immunological cross reactivity between serotypes 1 and 2 is easily demonstrated while cross reactivity between 2 and 3 may be detected but hardly between 1 and 3. The serological relationship between 1 and 2 can be epidemiologically demonstrated because presence of type 2 specific antibodies confers significant protection against type 1 infection [17].

Type 2 is more immunologically broad and that may be responsible for the fact that it is the first serotype to disappear during vaccination campaigns. The type 2 polioviruses were last seen in the world in 2002.

Genetic Differences Between the Polioviruses
Polioviruses, like other RNA viruses, have error-prone virus encoded RNA polymerase enzyme, which lacks proof reading activities. This results in rapid accumulation of mutations upon replication [18]

Epidemiologically, there are two categories of the poliovirus(WPV) which are also known as non- Sabin-like (NSL) and the Vaccine virus also known as Sabin –like (SL). These two categories are common to all the 3 serotypes. These two types of viruses are detected by intratypic differentiation tests

(ITD) which are based on one antigenic method, the polyclonal or monoclonal ELISA[19] and one molecular method which can enter the polymerase chain reaction (PCR) or RNA probe hybridization technique [20,21]. From this concordant non Sabin like ITD results are classified as wild, concordant Sabin-like as vaccine virus while any discordant results or sabin-like isolates lacking the two ITD tests are subjected to sequencing analysis of the major viral capsid surface protein, the VP1 [22].

In the sequence analysis, isolates with <1% difference from sabin-like virus are classified as sabin-like while isolates showing between 1%-15% difference in sequence analysis of the VP1 amino acids are classified as vaccine-derived poliovirus (VDPV).

Between the P1 wild prototype and the P1 sabin are 55 nucleotide changes. A change at position 480 of the amino acid sequence is responsible for the neurovirulence between the P1 wild and the Sabin1. There are 23 nucleotide changes between the P2 wild and the P2 Sabin while the neurovirulence is determined by a change in position 481 and one

other in the VP1. Only 11 nucleotide changes differentiate the wild P3 from its P3 sabin counterpart while the amino acid determining the neurovirulence is situated at position 472. The wild polioviruses irrespective of their serotypes consists of many genotypes, they are of high genetic diversity. They are highly transmissible and are usually highly neurovirulent. In contrast, the vaccine viruses originate from one original strain, are of low genetic diversity, low transmissibility and of very low neurovirulence unless on occasion when they have acquired the properties of VAPP or VDPV.

The oral polio vaccine (OPV) being a live-attenuated viruses vaccine behaves like the wild as regards evolution. The vaccine viruses may and usually do back mutate quite often during replication in human vaccinees [23]. When such vaccine virus strains back mutate and reacquire neurovirulence and transmissibility, the resultant virus is virtually wild-like [24]. Such OPV virus strains therefore cause paralytic poliomyelitis in susceptible vaccinees, although infrequently. This is called vaccine-associated paralytic poliomyelitis (VAPP). This is normal and occurs only very rarely. The chance of developing poliomyelitis without vaccination far much overweighs the possibility of developing a VAPP following vaccination. The option still remains therefore that vaccination is the best approach of preventing poliomyelitis.

The Polio Molecular Clock

Polioviruses are among the most rapidly evolving viruses known. The rapid evolution permits the pattern of poliovirus transmission to be followed with precision [25, 26]. Several factors combine to determine the overall rate of virus evolution. These include the replication error rates, the virus population size and growth rate, the frequency of genetic bottlenecks, the intensity of selective forces and the mechanism for genetic exchange [27]. Error rates for the poliovirus replicase have been estimated to be 10^{-4} to 10^{-5} per site per replication [28,29,30]. Nucleotide substitutions(90% of which in the coding region are synonymous codons) accumulate at overall rate of 10^{-2} substitutions per site per year and

at 3×10^{-2} substitutions per year at synonymous sites [25, 31, 32, 33, and 34].

Evolution rates are similar across serotypes and between wild and vaccine-derived polioviruses. Interestingly the bottlenecks driving the rapid evolution of polioviruses appear also to occur during replication in the human intestine in addition to that which happens during person-to-person transmission [35, 36]. Many poliovirus clinical isolates are recombinants [25, 35, 36]. Heterotrophic recombinants are frequently isolated from vaccinees given trivalent OPV [35, 37]. All wild polioviruses probably have a recent history of recombination because frequent genetic exchange with other species C enteroviruses and vaccine derived polioviruses appear to be typical of circulating polioviruses [25,38]. Crossover is most common in the non-capsid region, less common in the 5' nontranslated region and very rare nontranslated within the capsid region [39].Molecular clock data have been used to estimate the dates of the common ancestors to wild [25,26] and vaccine derived [34] polioviruses. For example it was possible to determine the date of receipt of an OPV dose that eventually gave rise to a type 2 vaccine derived poliovirus in a vaccinated Nigerian child that developed acute flaccid paralysis following vaccination with the OPV [40].

Circulating Vaccine-derived Poliovirus (cVDPV)

In recent times, another definition of poliovirus came into recognition. This is the circulating vaccine-derived poliovirus (cVDPV). These are revertant excreted recipient poliovirus vaccine strain OPV derived from the strain as a result of accumulating quantitative genetic change especially in the VPI capsid region. These viruses are usually related to the OPV strains from which they are derived but with significant genetic changes >1% difference. They are characterized by evidence of circulation and sustained person-to-person transmission and recombination with species C enteroviruses and other non-polio enteroviruses NPEV. cVDPVs behave very much like the wild type virus. They multiply very well at supra optimal temperature of 39.5° C. cVDVs outbreaks usually occur where the corresponding

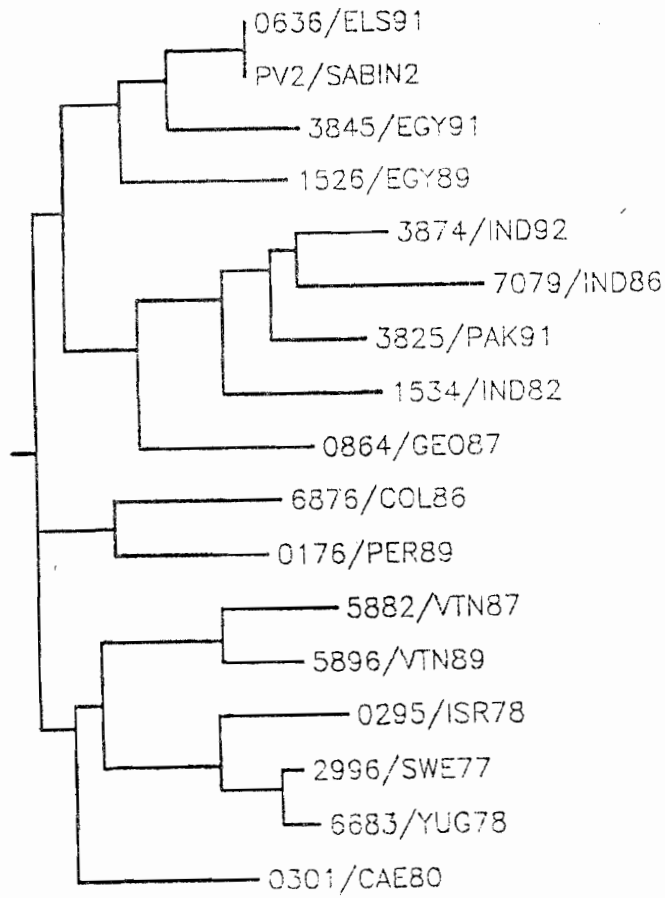


Fig. 2: Dendrogram summarizing sequence relatedness among 17 type 2 polioviruses across the interval of nucleotides 3295-3444(VPI/2A region)

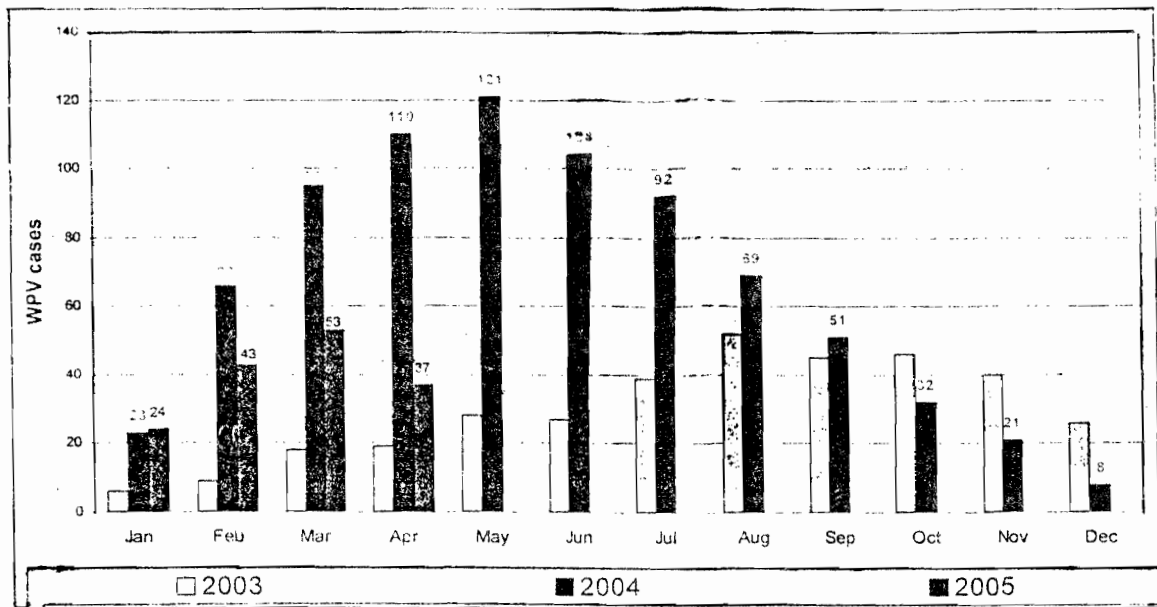


Fig. 3: Nigeria: Comparison of monthly WPV cases, 2003-16th April, 2005

serotypes have earlier been eradicated or eliminated. Factors favouring the appearance of cVDPVs include major gap in OPV coverage i.e. low vaccine coverage and other general environmental conditions favouring poliovirus spread.

Over the past 5 years there have been localized outbreaks of cVDPV in Hispaniola [24], the Philippines, [41] Madagascar, [42] and Nigeria [40]. Retrospective studies have also confirmed occurrences of cVDPV in Belarus, Poland and Egypt [43]. The appearance of cVDPV is posing a great threat to the polio eradication programme. Immunization managers must ensure in all countries that high polio vaccination coverage is maintained.

Virological Surveillance of Poliovirus

Efficient surveillance tools for wild polio and cVDPV are well developed. A global network of highly competent poliovirus laboratories has been established by WHO to apply these tools in tracking the virus and in guiding immunization campaigns [44]

Standard methods for poliovirus isolation in cultured cells [45] have been developed by use of recombinant murine cells expressing the poliovirus receptor [46]. Sero differentiation of the three poliovirus serotypes has been extended through the development of cross-absorbed antisera that distinguish OPV-derived strains from wild polio virus isolates. The most definite method is however based upon the sequence differences among poliovirus genomes [47]. Sets of highly specific nucleic acid probes [20] and polymerase chain reaction (PCR) primers [21] have been developed for routine identification of poliovirus isolates. By far the most powerful approach is comparative genomic sequencing [47] Sequence relationships can be efficiently summarized in the form of dendograms (Fig 2). With the application of genomic sequencing and molecular epidemiological approaches, it is now possible to determine the source of imported viruses, follow the pathway of virus transmission, monitor the progress of control activities, identify reservoirs sustaining virus transmission and develop molecular reagents for rapid detection of wild poliovirus.

Wild Poliovirus in Nigeria

Nigeria remains one of the six countries of the world where the wild poliovirus is still endemic. The virus is still very much in circulation especially in the northern band of states where routine immunization and mass immunization is still not efficient enough to stop the transmission of the virus. Last year alone, Nigeria topped the list of countries of the world in reporting the largest number of wild poliovirus with a total of 867 [48]. In 2005, Nigeria accounts for 35% of the global wild poliovirus case count and 92% of the cases in Africa since the beginning of 2005. Between January 2nd and 16th April 2005, a total of 157 wild poliovirus cases have been reported from 18 states of the federation (Fig 3). Of the 18 infected states, 7 states of the North Western Zone account for 68% of the total case count. 78% of the wild poliovirus cases are children below 3 years of age and 71% have received less than 3 doses of OPV. Nigeria has not only remained the main reservoir of the wild poliovirus, poliovirus originating from Nigeria have been imported to many countries of the world like Ghana, Burkina Faso, Chad, Sudan, Ethiopia, Saudi Arabia, Malawi, Indonesia and lately Yemen.

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