Overview of the WHO Polio Laboratory Network in the African Region

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A plan of action was formulated in 1989 detailing laboratory support for the global eradication of poliomyelitis. Its function was to describe the activities needed to establish a three-tiered global network of polio laboratories, each with well-defined responsibilities. Today, at all three levels of the network (national, regional and global specialized), laboratories work together in the largest coordinated public health laboratory network.

The WHO Polio Laboratory Network (LabNet) in the African Region plays a critical role in the global Polio Eradication Initiative (PEI). Timely, accurate laboratory results drive public health action and help shape policies. The mission of LabNet contributes to improving the quality of laboratory services for effective acute flaccid paralysis (AFP) surveillance.

The network that was established in 1993 and provides support to 47 African Region and 1 Eastern and Mediterranean Region countries, with 16 polio laboratories, namely: Algeria, Cameroon, Central African Republic, Côte d’Ivoire, Democratic Republic of the Congo, Ethiopia, Ghana, Nigeria (with two labs), Kenya, Madagascar, Senegal, South Africa, Uganda, Zambia and Zimbabwe, assigned to perform laboratory diagnosis of polioviruses. The three laboratories in Ghana, Central African Republic and South Africa are the regional reference laboratories. The global reference laboratory at CDC Atlanta supports sequencing laboratories in South Africa and in Ghana by further analysing query sequencing and performing a regional level function for the Nigeria and Cameroon laboratories.

The eradication of poliomyelitis will be accomplished only when these laboratories provide convincing diagnostic evidence of the absence of wild poliovirus infections in humans and prolonged circulation in the environment. In addition, because these laboratories store specimens from AFP cases and wild polioviruses isolates, containment of these materials and viruses remains one of the laboratories’ key responsibilities as a pre-requisite for certifying Africa free of polio.

From 2006, LabNet made changes to the laboratory diagnostics protocols and this has improved laboratory performance and reduced the results’ turnaround time dramatically. These changes have necessitated strengthening laboratory quality control and quality assurance systems through the implementation of new algorithms.

There is growing recognition that the quality of LabNet serves as catalyst for the PEI by providing timely results. This article outlines some of the progress, performance and critical support provided by LabNet towards achieving the polio eradication target in Africa.

Methodology

The laboratory performance indicators listed in Table 1 provide information about the capability and the capacity of any laboratory to detect, identify and promptly report wild polioviruses, vaccine-derived polioviruses (VDPV)
and Sabin viruses that may be present in clinical and environmental specimens. These laboratory indicators can further indicate if there is a need for learning opportunities, serve as a mechanism for identifying resources and training, and can be a measure of the progress of a laboratory.

**Laboratory performance**

From 2010 to August 2014, 187,837 samples were received by LabNet for viral isolation (Figure 1) with Nigeria’s Ibadan laboratory processing approximately 50,000 and 50% of labs processing more than 10,000 samples. All laboratories managed to report the isolation results within the 14 days’ turnaround time.

In the same period, 18,097 isolates were received for intratypic differentiation and two laboratories received over 3,000 isolates. The ITD results were reported within the expected time of seven days and the 80% target was reached by all 16 laboratories.

**Virus isolation**

The faecal specimens of AFP cases were processed at the national polio laboratories for virus isolation as per the WHO laboratory manual (version 4). Briefly, tube cultures of cell lines were inoculated with 0.2 ml of specimen extract and incubated in the stationary sloped (50°) position at 360°C. Cultures were monitored daily using standard or inverted microscopes for the appearance of cytopathic effect (CPE). All cell lines with characteristics of enterovirus CPE were stored at 20°C for a second passage in a tube containing 2 ml of medium. Second passage material with CPE were reported as suspected poliovirus and forwarded to regional laboratories for intratypic differentiation (ITD).

**Intratypic differentiation**

Intratypic differentiation assay is used to screen out isolates that are wild polioviruses from those that are vaccine strains. All viral isolates that had shown CPE (infected with virus) in L20B or human rhabdomyosarcoma (RD) cell line were tested using an rRT-PCR (real-time assay) that included separate reactions with specific poliovirus strains, and specific serotype 1, 2 and 3 polioviruses and further screening for vaccine-derived polioviruses (VDPVs).

**Table 1. Laboratory performance indicators**

<table>
<thead>
<tr>
<th>Laboratory performance indicators</th>
<th>Target</th>
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<tbody>
<tr>
<td>Results of all AFP specimens reported within 14 days</td>
<td>80%</td>
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<tr>
<td>Intratypic differentiation (ITD) test results of poliovirus isolates reported within seven days</td>
<td>80%</td>
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<tr>
<td>Suspected poliovirus isolates from AFP cases and contacts are forwarded for ITD within seven days, if applicable</td>
<td>80%</td>
</tr>
<tr>
<td>Non-Sabin-like poliovirus and ITD-discordant isolates of AFP cases, contacts and other sources are referred for sequencing within seven days of detection, if applicable</td>
<td>80%</td>
</tr>
<tr>
<td>Accuracy of identification of polioviruses among all isolates obtained in cell lines</td>
<td>90%</td>
</tr>
<tr>
<td>Accuracy of poliovirus serotyping and intratypic differentiation</td>
<td>90%</td>
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</tbody>
</table>

**Figure 1. Stool samples received by 16 polio laboratories**

![Figure 1](image1)

**Figure 2. Isolates received by 16 polio laboratories**

![Figure 2](image2)
Results

Samples reported as suspected polioviruses were tested by ITD assay and were identified as wild polioviruses and discordant Sabins. All wild polioviruses and discordant Sabins were further characterized by the sequencing method.

Of the tested isolates, 17,560 isolates identified the following poliovirus strains: wild 1 was identified on 3,089 isolates, 5,147 were Sabin 1, 2 were VDPV1, no wild 2 was identified, 3,073 were Sabin 2, 115 were VDPV2, 346 were wild 3, 5,599 were Sabin 3, 2 were VDPV3 and 187 isolates were identified as non-polio enteroviruses (NPEV) (Figure 3).

All 16 laboratories staff have been trained and are fully competent in performing the ITD assay. This has reduced the turnaround time of results to be reported to the programme. In addition, the sequence capacity has been expanded in order to increase the number of laboratories capable of analysing viruses by sequencing. All laboratories are fully accredited.

Discussion

LabNet is made up of 16 laboratories that perform high-level quality assurance systems to ensure the production of excellent, timely and accurate results. The detection of different poliovirus strains in the Region by LabNet within set turnaround times is an indication of a well-managed network. Successful implementation of new diagnostic technology reduced reporting results times by around 50%, which has greatly assisted the PEI programme.

The results produced by the laboratories have been used to answer epidemiological questions regarding the likely location of endemic virus reservoirs, patterns of virus transmission or source of imported strains and are contributing to the progress towards the eradication of polioviruses.

At the country level, the integration of laboratory networks such polio, measles, yellow fever, rotavirus and influenza has taken root with resultant shared technologies, shared human resources, joint planning and support of missions. This will ensure that the investment in polio eradication provides public health dividends for years to come.

Conclusion

LabNet has implemented and maintained high standards of performance and set the stage for laboratory integration in Africa. The network has significantly reduced the results’ turnaround time by more than 50%. Genetic sequence information generated suggests regional progress in reducing the wild poliovirus reservoir as evidenced by a reduction in the number of circulating viruses.

References