Guidelines for Implementation of Laboratory Containment of Wild Poliovirus

Laboratory Survey and National Inventory

May 2000
EUROPEAN HEALTH21 TARGET 7

REDUCING COMMUNICABLE DISEASES

By the year 2020, the adverse health effects of communicable diseases should be substantially diminished through systematically applied programmes to eradicate, eliminate or control infectious diseases of public health importance

(Adopted by the WHO Regional Committee for Europe at its forty-eighth session, Copenhagen, September 1998)

Keywords

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CERTIFICATION
CONTAINMENT OF BIOHAZARDS STANDARDS
SPECIMEN HANDLING STANDARDS
LABORATORY INFECTION – prevention and control
POLIOVIRUS PATHOGENICITY
SAFETY MANAGEMENT
EUROPE
Acknowledgements

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Executive Summary

This document provides guidelines for implementation of the Action Plan for Laboratory Containment of Wild Polioviruses in the WHO European Region. Implementation of the activities outlined in the document will insure that all locations of wild poliovirus (or potentially infectious material) will be disposed of or are documented and under proper containment.

In 1988, the World Health Assembly adopted the resolution calling for global eradication of poliomyelitis by the year 2000. Three WHO regions have not isolated wild poliovirus for >1 year despite excellent surveillance. In the European Region, the last case of poliomyelitis associated with isolation of wild poliovirus had onset on 26 November 1998, and more than one year has passed without additional virus isolation. Therefore, the goal of poliomyelitis eradication appears within reach in the European Region. Rapid progress toward eradication is reported from the other regions as well.

By the end of 2000, or shortly thereafter, poliovirus transmission will be interrupted globally, the only potential source of wild polioviruses, will be the biomedical laboratories, including diagnostic, research, teaching, and vaccine production laboratories. Therefore it is imperative that laboratories prepare now for the time when: 1) poliovirus transmission has ceased; and 2) poliomyelitis vaccination will be stopped. The activities outlined in the document should greatly reduce the potential risk of reintroduction of poliovirus into the population from a laboratory. A reintroduction of virus would constitute a public health emergency of global proportions.

In May 1999, the Fifty-second World Health Assembly reaffirmed the commitment of the world community for global poliomyelitis eradication by the end of the year 2000, and urged poliomyelitis-free Member States to begin, in collaboration with WHO, the process leading to the laboratory containment of polioviruses. To this end, the WHO Regional Office for Europe has drafted a Regional Plan of Action for the Containment of Wild Polioviruses.

Before a WHO region can be certified by the Regional Commission for the Certification of Polio Eradication as poliomyelitis-free, all countries of the region need to survey and prepare an inventory (documented in a national registry) of biomedical laboratories that store wild poliovirus infectious and/or potentially infectious materials. The laboratories are encouraged to destroy these materials or send them to a national or international repository for storage. Laboratories wishing to retain the materials must provide detailed documentation and comply with the appropriate biosafety requirements for storage and handling. Laboratories to be surveyed include not only those associated with traditional poliovirus laboratory functions but also laboratories in other sectors outside the Ministry of Health that could have wild polioviruses and/or potentially infectious materials. Although the probability of a laboratory-associated poliovirus infection is small, the consequences of such infection (and reintroduction of virus into the population) grow greater with time after stopping vaccination.

The purpose of containment, a phased approach depending on progress toward eradication, the certification process, and the plans for stopping poliomyelitis vaccination is explained in the WHO Global Action Plan for Laboratory Containment of Wild Polioviruses.
The European Regional Guidelines outline the activities necessary for the implementation of the pre-eradication phase of the Action Plan. The two major activities are:

- a national survey of all medical/biological laboratories that might possess wild poliovirus infectious and/or potentially infectious materials (laboratories are encouraged to dispose of these materials); and
- a national inventory system of laboratories that wish to retain such materials.

The National Survey is hierarchical, beginning with notification by WHO to the highest authority of each country or the Ministry of Health which, in turn, should appoint a Focal Person/Group of National Coordination (National Coordinator) for Containment to assume operational responsibility for the national containment process. The national coordinator will draft the National Plan for Containment and will assume responsibility for the national survey of laboratories. This process includes contacting agencies and institutions who, in turn, will ask laboratories under their jurisdiction to initiate and document the search for poliovirus infectious and/or potentially infectious materials. Because many of the laboratories are outside the health sector (i.e., not under the control or supervision of the Ministry of Health), successful completion of the survey will require the cooperation of other ministries, including Education, Defense, Agriculture and Environment.

A National Inventory System will be established based on data generated by the National Survey. Each laboratory retaining wild poliovirus infectious or potentially infectious material will submit a detailed inventory of all such materials to its parent Agency/Institution, which in turn will create an Agency/Institution Inventory. Data from all laboratories listed on the latter will be included in the National Inventory, maintained centrally by each country. Summary data from the National Inventory are submitted to the WHO Regional Office to be included in the Regional Inventory, which, in turn, will forward the appropriate data to the WHO Global Inventory.

The subsequent phases of the Action Plan call for increasing levels of biosafety for laboratories wishing to retain infectious and potentially infectious materials.

Completion of National Inventories for each country in the region is a prerequisite for certification of the region as free-of-poliomyelitis, and close collaboration will be needed between the national containment coordinator and the national committee for the certification of poliomyelitis eradication.
Introduction

Once poliomyelitis is eradicated, the laboratories of the world will be the only remaining source of the virus. Safe handling and, ultimately, maximum containment of poliovirus and potentially infectious materials in the laboratory is crucial.

Until now, poliovirus biosafety concerns have been minimal. Universal immunization with inactivated poliomyelitis vaccine (IPV) or oral poliomyelitis vaccine (OPV) has reduced the risk of disease for laboratory workers and the general public. Current day technologies and biosafety practices have further reduced those risks, as well as poliovirus contamination of the environment.

The probability of a laboratory-associated poliovirus infection is small, but the consequences of an infection grow greater with time. A chance reintroduction of wild polioviruses from the laboratory into the community after cessation of transmission presents a threat to poliomyelitis eradication. A chance reintroduction of wild poliovirus after cessation of immunization presents a threat to public health of global proportions.

The world now faces the formidable, but not insurmountable, challenge of locating the many laboratories that have wild poliovirus infectious, or potentially infectious materials, and ensuring that they are adequately contained in the laboratory, rendered non-infectious, or destroyed. The required action consists of three phases, which are linked to the major eradication objectives.

Pre-certification of Region as poliomyelitis-free

Time frame: To begin immediately.

Activity: Safe handling of wild poliovirus infectious or potentially infectious materials (BSL-2/polio).

This phase covers the period when wild poliovirus is no longer circulating in the Region. Three tasks are critical:

1. Nations must identify and develop an inventory of laboratories that retain wild poliovirus infectious or potentially infectious materials.
2. Laboratories must institute enhanced biosafety level–2 (BSL-2/polio) procedures for safe handling of all such infectious or potentially infectious materials.
3. Nations must decide how each laboratory on the inventory will deal with implementation of biosafety requirement for post-global poliomyelitis eradication phase.

Post-global poliomyelitis eradication

Time frame: To begin within one year after detection of the last wild poliovirus anywhere in the world.

This phase, post global eradication, begins one year after detection of the last wild poliovirus anywhere in the world, time when the probability is high that all human transmission has ceased. All laboratories possessing wild poliovirus infectious materials or potentially infectious materials must elect one or more of the following three options:

1. implement containment BSL-3/polio procedures; or
2. transfer wild poliovirus infectious and potentially infectious materials to WHO designated repositories; or
3. render such materials non-infectious, or destroy them, under appropriate conditions.

All required biosafety actions are to be implemented and documented as complete before global certification of poliomyelitis eradication can be considered.

**Post-OPV immunization**

**Time frame:** To begin when OPV immunization stops

**Activity:** Maximum containment (BSL-4) of wild poliovirus infectious and potentially infectious materials and high containment (BSL-3/polio) of OPV and OPV–derived viruses.

This phase, post-OPV immunization, begins with the worldwide cessation of OPV administration and the subsequent rapid increase of non-immune susceptible children. The biosafety requirements for wild poliovirus infectious and potentially infectious materials increase from BSL-3/polio to BSL-4, consistent with the increased consequences of inadvertent transmission of wild poliovirus from the laboratory to the community. Biosafety requirements for OPV and OPV–derived viruses increase from BSL-2/polio to BSL-3/polio to prevent reintroduction and potential circulation of these viruses in unimmunized populations. Procedures will be developed to control or destroy unused OPV in clinics, immunization centres, physician’s offices, and other sites.

For further details concerning these phases, please consult the **Action Plan for Laboratory Containment of Wild Polioviruses**.

**Evidence for laboratory-associated infections**

From 1941 to 1976 a total of 12 laboratory-associated poliomyelitis cases including two deaths, were recorded. Most cases occurred in the pre-vaccine era and before the advent of cell culture. The paucity of reports of laboratory-associated poliomyelitis since vaccines were introduced testifies to the effectiveness of vaccines and the vast improvement of laboratory facilities, technologies, and procedures. By inference, poliovirus infections in the absence of clinical disease would also be expected to be rare among laboratory workers.

Despite the advances in biosafety over the past 40 years, recent evidence indicates that the potential nevertheless exists for transmission of poliovirus from the laboratory to the community. In 1992, a wild-type 1 strain used for IPV production was documented as being transmitted from a worker in a vaccine production facility to his young child.
Although IPV is highly effective in preventing disease, its use cannot be assumed to prevent silent infection among laboratory workers. Using the current OPV to provide a more effective, but still incomplete, barrier to infections may not be an option. At some point after eradication, OPV may be prohibited worldwide to avoid the potential spread of vaccine-derived virus in the general population.

**Defining terms**

**Definitions of poliovirus**

Polioviruses are defined by standard neutralization tests with specific antisera. The three poliovirus serotypes form a unique genetic group of human enteroviruses that initiate infection by binding to a specific cellular receptor (PVR:CD155). Wild polioviruses have the capacity to circulate indefinitely within susceptible human populations. Important determinants of the attenuation phenotype reside in the capsid regions of OPV strains, and these determinants are not known to occur in the capsid sequences of wild polioviruses. Candidate attenuated strains that are not approved for use in oral poliomyelitis vaccines by national control authorities are regarded as wild polioviruses.

Definitions of poliovirus are presented in Box 1.

<table>
<thead>
<tr>
<th>Box 1 – Definitions of poliovirus*</th>
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</thead>
<tbody>
<tr>
<td><strong>Polioviruses:</strong> human enteroviruses that exist as three well-defined serotypes, which infect cells via a specific receptor (PVR:CD155).</td>
</tr>
<tr>
<td><strong>Wild polioviruses:</strong> field isolates and reference strains derived from polioviruses known or believed to have circulated persistently in the community.</td>
</tr>
<tr>
<td><strong>Oral poliovirus vaccine strains:</strong> attenuated polioviruses approved for use in oral vaccines by national control authorities.</td>
</tr>
<tr>
<td><strong>Vaccine-derived polioviruses:</strong> progeny of approved oral poliovirus vaccine strains.</td>
</tr>
</tbody>
</table>

* From the WHO Global Action Plan for Laboratory Containment of Wild Polioviruses.

**Wild poliovirus infectious materials**

Wild poliovirus may be present in faeces and throat specimens; less commonly in blood; and rarely in cerebrospinal fluids from patients with non-paralytic as well as paralytic infections. In fatal infections, wild poliovirus may be present in faeces, intestinal contents, lymph nodes, brain tissue, and spinal cord tissue. All such clinical materials from persons known or suspected to be infected are defined as infectious, even though the presence of virus may not have been confirmed.

Other infectious materials are wild poliovirus isolates, reference strains, and all products of the laboratory that meet the definitions of wild poliovirus (Box 1). Also included are environmental sewage or water samples known or suspected to be contaminated, infected laboratory animals, and materials from infected animals. Definitions and examples of infectious materials are presented in Box 2 below.
## Box 2 – Definitions and examples of wild poliovirus infectious materials

### Infectious clinical materials:
- All clinical and investigative materials from confirmed or suspected cases of poliomyelitis.

**Examples:** Specimens from suspected or confirmed poliomyelitis cases collected for laboratory studies:
- throat, faecal, blood, cerebrospinal fluid, autopsy and biopsy.

### Infectious research materials:
- All poliovirus derivatives produced in the laboratory that have capsid sequences derived from wild polioviruses.
- Full-length poliovirus RNA or cDNA containing capsid sequences derived from wild poliovirus.
- Cells persistently infected with poliovirus strains whose capsid sequences are derived from wild poliovirus.

**Examples:**
- Stocks of wild viruses:
  - Prototype strains used as controls
  - Isolates
  - Proficiency test panels
  - Seeds for inactivated vaccines
- Materials with wild poliovirus capsid sequences:
  - Poliovirus derivates
  - Full length poliovirus RNA or cDNA
  - Infected cells

### Infectious animals:
- Any experimental animal infected with a strain containing capsid sequences derived from a wild poliovirus, especially CD155 transgenic mice infected with wild poliovirus.
- Specimens from laboratory animals infected with wild virus (non-human primates, transgenic mice, etc.)

### Infectious environmental materials:
- Sewage or water samples known or suspected to contain wild polioviruses.

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**Wild poliovirus potentially infectious materials**

The search for laboratories containing potentially infectious materials will include biomedical laboratories inside as well as outside of the health sector. A thorough search of laboratories unrelated to poliomyelitis work is crucial for identifying potentially infectious materials that would otherwise be missed. Potentially infectious materials include any clinical and environmental specimen compatible with the potential presence of poliovirus collected for any diagnostic or research purposes at a time and in a geographic area where wild poliovirus was circulating in an endemic or epidemic manner. All such clinical and environmental materials must be carefully evaluated for potential infectivity.

Each collection must be assessed to determine the likelihood of the presence of wild polioviruses, based on treatment and storage history, the area of origin, the year collected, the time of the last indigenous wild poliovirus isolates in the area, and the type of specimen. Frozen stool samples from young children during endemic periods would have the highest probability of containing infectious polioviruses.
Routinely collected serum specimens and cerebrospinal fluids are not likely to contain sufficient levels (if any) of poliovirus to cause infection and are not considered potentially infectious (unless obtained from known or suspected cases of poliomyelitis).

The manner in which potentially infectious materials have been treated and stored also affects whether a material should be considered potentially infectious or not. Box 3 details these conditions.

Many types of materials should be included in the search for potentially infectious materials. They include not only those collected from patients but also products of laboratory research techniques and specimens from certain environmental investigations. Box 3 details some of the types of materials that should be considered. Laboratories should be asked about any such materials that they contain and then do further investigation to determine the conditions of these materials collection and storage.

**Box 3 – Definition and examples of potentially infectious laboratory materials**

| Potentially infectious laboratory materials:* | clinical materials particularly faeces and throat swabs; autopsy specimens; and environmental samples collected for any purpose at a time and in a geographical area where wild poliovirus was known or suspected to be present (see Annex 3 for list of countries and last known date of poliovirus circulation). |
| Examples: | |
| Clinical materials* | Faeces |
| Autopsy specimens (unfixed)* | Faeces and intestinal contents; Lymph nodes; Brain tissue; Spinal cord tissue |
| Laboratory products* | Untyped enterovirus-like; Cell culture isolates; Undifferentiated poliovirus isolates |
| Environmental sewage and untreated water samples* | |

*Conditions excluding materials as potentially infectious

- clinical materials stored without refrigeration for three months or more
- refrigerated for one year or more (NOTE: frozen materials ARE potentially infectious)
- heat inactivated at 50°C
- treated with a disinfectant known to inactivate polioviruses
  - 3% formaldehyde, 1N HCL, or free residual chlorine at 0.3 – 0.5 ppm are examples of effective disinfectants (NOTE: alcohol (70%), 5% Lysol, 1% quaternary ammonium compounds, ether, or deoxycholate are NOT effective disinfectants)
- tested and found negative for the presence of enteroviruses by:
  - cell culture
  - PCR/gene amplification
National containment activities

Preparations

Successful containment and documentation of wild poliovirus infectious and potentially infectious materials within a nation’s laboratories will require coordination between various branches of government and the private sector. The level of coordination required will differ from country to country dependent on the extent of that nation’s laboratory capacity. In order to coordinate all of the different institutions potentially involved with containment, it is recommended that each country appoint a Focal Person (National Containment Coordinator)/Group for Containment. This coordinator will be responsible for creating a National Plan for Containment and insuring that all activities called for are implemented in time to be ready for Regional Certification as “Polio-Free”. By this time, each nation should have submitted all documentation to the WHO Regional Office.

Soon after the National Containment Coordinator has been appointed, this person/group should meet with the National Committee to review and use these Guidelines to create a National Plan for Containment. The National Containment Coordinator/Group should give careful consideration to determining which Agencies/Institutions and laboratories should be included in the survey.

Once the National Plan for Containment has been finalized, the National Containment Coordinator should begin to implement the survey of all potential laboratories. Depending on national circumstances, this could involve sending letters and the appropriate forms to each Agency/Institution or laboratory explaining the goals of containment and requesting their cooperation to carry out the survey of individual laboratories. The findings of the survey should be documented (see attachment of sample forms). After completion of the survey, the director of the laboratory should sign the documents before sending these back to the requesting Agency/Institution who in turn will compile the information of all laboratories under their jurisdiction and forward this information to the National Containment Coordinator.

The National Containment Coordinator will be responsible for reviewing completed forms submitted from each agency/institution and laboratory and maintain an inventory of all laboratories surveyed. The National Containment Coordinator will also decide if a material is to be considered infectious, potentially infectious, or non-infectious. The disposition of materials categorized as infectious and/or potentially infectious is encouraged. If materials are to be preserved, for whatever reason, they need to be stored under increasing levels of biosafety containment as described in the “Introduction” section of this document.

Two following major activities need to be specified in detail in the National Plan for Containment:

- a national survey for all biomedical laboratories that might possess wild poliovirus infectious and/or potentially infectious materials;
- a national inventory system for laboratories that retain such materials.

Detailed instructions, guidelines, with the appropriate forms should be forwarded from the national level to each agency/institution that will distribute the package of information to laboratories under their jurisdiction.
After receiving the information package, each laboratory will perform a search of their working and storage spaces for poliovirus infectious and/or potentially infectious materials and document the search for such materials using the appropriate inventory forms, and adhering to the deadlines provided. The laboratories are encouraged to destroy infectious and potentially infectious materials if there are no compelling reasons to retain these. After completion of the search, the laboratory director will submit the completed inventory forms to their parent Agency/Institution, who will in turn compile an Agency/Institution Inventory. These inventories in the laboratory and the parent Agency/Institution should be kept on active file and regularly updated to reflect any changes in the status of materials. The forms to be used by laboratories can be found in Annex 3.

**Global inventory**

Data to be submitted by each agency/institution to the National Inventory will include: documentation of laboratory search for wild poliovirus infectious and/or potentially infectious materials; names and addresses of laboratories that retain such materials; type of laboratory; nature of materials; dates of most recent information; and monitoring of the accuracy of reports. The form to be used by agencies/institutions to be submitted to the National Inventory is contained in Annex 2.

The National Inventory will retain on active file the completed reports from the survey which will be used to prepare summary reports. The National Inventory will include documentation of the laboratory search, names of agencies/institutions that contain materials, number of laboratories according to types and whether containing infectious, potentially infectious materials, or both, and dates of most recent reports from the agencies/institutions. The form to be completed by the National Inventory is contained in Annex 1.

A summary report of the data available in the National Inventory should be provided to the WHO Regional Inventory and the National Committee for the Certification of the Eradication of Poliomyelitis. Completion of a National Inventory is a prerequisite for certification of a country as poliomyelitis-free.

One year after detection of the last wild poliovirus, WHO will request countries to notify all agencies/institutions on the National Inventory to instruct laboratories to implement procedures for the next phase of the Global Plan for Laboratory Containment of Wild Polioviruses as outlined in the “Introduction” section of this document.

Potential indicators to monitor the progress of a National Plan for Containment are listed in Box 4.

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### Box 4 – Potential indicators to monitor progress of containment activities

| National Level: | 1. Number and type of different agencies responding nationwide  
Number of potential agencies contacted  
2. Number of found materials with a designated plan (destroyed, moved, etc.)  
Number of found materials |
|----------------|--------------------------------------------------------------------------|
| Agency/Institution level: | 1. Number of laboratory complete inventory forms received  
Number of laboratory inventory forms sent |
| Laboratory level: | 1. Number of refrigerators/freezers searched  
Total number of refrigerators/freezers in laboratory |
Box 5 – Global scheme to contain laboratory wild poliovirus infectious and/or potentially infectious materials

Who provides Guidelines to Member States and requests them to begin a national survey

Countries (MOH) appoint a national coordinator/group for containment to organized and monitor containment

National coordinator/group and national committee create the National Plan for Containment

National coordinator/group requests agencies/institutions identified in the plan to survey all biomedical laboratories under their jurisdiction

Agencies/institutions request laboratories to search facilities

Laboratories prepare inventory*

No poliovirus any more

Wild poliovirus stated stored

Statement to the National Committee

GLOBAL INVENTORY
Compilation of Regional Inventories

REGIONAL INVENTORY
Search data provided by Member States

NATIONAL INVENTORY
Data from agencies and institutions with laboratories containing wild poliovirus infectious and potentially infectious materials

AGENCY/INSTITUTIONAL INVENTORY
Lists from laboratories containing wild poliovirus infectious materials

LABORATORY INVENTORY**
Current lists of all wild poliovirus infectious materials including location and storage condition

* Laboratories are encouraged to effect disposal of wild polioviruses infectious and/or potentially infectious materials no longer needed by the laboratory following the recommendations mentioned in attachment V and certify it.

** Laboratories that wish retain infectious and potentially infectious materials should make laboratory inventory of such materials.
A suggested schedule to organize a National Plan for Containment is listed below:

<table>
<thead>
<tr>
<th>Activity</th>
<th>October/December</th>
<th>January/March</th>
<th>April/June</th>
<th>July/September</th>
<th>October/December</th>
<th>January/March</th>
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<th>July/September</th>
<th>October/December</th>
<th>January/March</th>
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<tbody>
<tr>
<td>1. Guidelines and GAP* sent by WHO to ministries of health of Member States (guidelines and action plan are translated).</td>
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<td>2. National coordinator/group for containment is appointed.</td>
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<td>3. National coordinator/group:</td>
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<td>b. creates national plan for containment;</td>
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<td>c. lists agencies/institutions and laboratories to include in the national search;</td>
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<td>d. meets national certification committee.</td>
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<td>4. Ministry of health sends letters to all agencies/institutions asking for their cooperation with the containment effort. The letters should include:</td>
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<td>a. explanation and instructions regarding containment;</td>
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<td>b. forms to perform the laboratory search and inventory;</td>
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<td>c. a deadline for receiving completed documents.</td>
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<td>5. Agencies/institutions notify laboratories under their jurisdiction to begin the laboratory inventory using the flow chart and forms provided. Laboratories should:</td>
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<td>a. decide on the future of the materials and document any material destroyed;</td>
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<td>b. fill out form to record all materials retained;</td>
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<td>c. send the completed and signed form to parent agency/institution.</td>
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<td>6. Agencies/institutions follow up laboratories to ensure replies are received from all recipients.</td>
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<tr>
<td>7. National coordinator/group follows up defaulters and visits larger laboratories and those with the most material classified as potentially infectious</td>
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<td>8. National committee meets national coordinator/group to review documents and prepare report to be sent to WHO Regional Office.</td>
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</table>

National survey of laboratories that might possess wild poliovirus infectious and/or potentially infectious materials

The purpose of the National Search is:

- to establish a National Inventory of laboratories that contain such materials;
- to acquaint all biomedical laboratories with the global/Regional/National Action Plans for Containment;
- to destroy all wild poliovirus infectious and/or potentially infectious materials no longer needed by the laboratory;
- to ensure safe handling of all such materials.

Diagnostic and public health laboratories keep poliovirus isolates and clinical specimens for documentation of past investigations of endemic, epidemic or imported cases of poliomyelitis. Some maintain multiple virus strains for test controls or reference purposes. Educational institutions may have wild polioviruses for teaching exercises. Virus research laboratories often retain poliovirus stocks or infectious materials for studies on the biologic, biochemical, or genetic properties of the virus. Other research laboratories store potentially infectious materials as documentation of completed studies or for future studies. Some environmental laboratories retain contaminated materials, wild poliovirus reference strains, or wild virus to use for tests on the effectiveness of virocidal compounds. Vaccine producers have wild strains for the production of IPV or to test the quality of OPV. National Control Laboratories may have similar strains.

Venues for identifying laboratories with wild poliovirus or potentially infectious materials include government sources, national laboratory registries, accrediting bodies, professional organizations, and national and institutional biosafety infrastructures.

Most challenging to identify are laboratories with potentially infectious clinical, epidemiological, research, or environmental specimens collected for other purposes at a time and in a geographical area of wild poliovirus endemicity. This implies that any laboratory, regardless of size or focus, cannot be assumed automatically to be free of wild poliovirus potentially infectious materials. Therefore, the search for potentially infectious materials must include all biomedical laboratories.

Box 6 lists potential Agencies/Institutions and laboratories that will need to be considered to include in the National Plan for Containment. There also may be other laboratories not found on this list and which should be considered by the National coordinator/group in development of the National Plan for Containment.
Box 6 – Agencies/institutions and laboratories that might possess wild poliovirus infectious and/or potentially infectious materials

<table>
<thead>
<tr>
<th>Category</th>
<th>Subcategory</th>
<th>Laboratories that might possess wild poliovirus infectious and/or potentially infectious materials</th>
</tr>
</thead>
<tbody>
<tr>
<td>Biological Control Agencies</td>
<td>National/Provincial</td>
<td>Microbiology laboratories* (bacteriology, mycology, parasitology, and virology)</td>
</tr>
<tr>
<td>Biomedical Research Institutions</td>
<td>National/Provincial/Commercial/Non-profit</td>
<td>Pathology laboratories* Haematology laboratories* Neurology laboratories* Gastroenterology laboratories* Nutrition laboratories* Environmental laboratories*</td>
</tr>
<tr>
<td>Culture Collections</td>
<td>National/Institutional</td>
<td>*includes control, diagnostic, production, research, and teaching laboratories</td>
</tr>
<tr>
<td>Environmental Agencies</td>
<td>National/Provincial/Local</td>
<td>Microbiology laboratories* Pathology laboratories* Haematology laboratories* Neurology laboratories* Gastroenterology laboratories* Nutrition laboratories* Environmental laboratories*</td>
</tr>
<tr>
<td>Hospitals/Clinics</td>
<td></td>
<td>*includes control, diagnostic, production, research, and teaching laboratories</td>
</tr>
<tr>
<td>Military Agencies</td>
<td>Health/Research</td>
<td>*includes control, diagnostic, production, research, and teaching laboratories</td>
</tr>
<tr>
<td>Producers</td>
<td>Biologics/Vaccines</td>
<td>*includes control, diagnostic, production, research, and teaching laboratories</td>
</tr>
<tr>
<td>Public Health Agencies</td>
<td>National/Provincial/Local</td>
<td>*includes control, diagnostic, production, research, and teaching laboratories</td>
</tr>
<tr>
<td>Universities</td>
<td>Food Safety</td>
<td>*includes control, diagnostic, production, research, and teaching laboratories</td>
</tr>
<tr>
<td>Agencies unique to country Structures</td>
<td></td>
<td>*includes control, diagnostic, production, research, and teaching laboratories</td>
</tr>
</tbody>
</table>

*From the WHO Global Action Plan for Laboratory Containment of Wild Polioviruses.

The National Survey forms to be submitted to WHO along with all the supporting documentation can be found in Annex 1.
Agency/institution inventory of laboratories that contain wild poliovirus infectious and/or potentially infectious materials

Agencies/institutions that have jurisdiction over laboratories that might contain wild poliovirus infectious and potentially infectious materials should establish an Agency/Institutional Inventory. Such Inventories should be located at the highest organizational level. Smaller countries may elect to omit Agency/Institutional Inventories, operating only a National Inventory. Although a single Inventory in such countries is encouraged, care must be taken to it includes all the relevant information.

Critical data elements of the Agency/Institutional Inventory include at a minimum the following information:

- Documentation of search (numbers of laboratories searched)
- Names and addresses of laboratories that contain infectious or potentially infectious wild poliovirus materials
- Types of laboratories (control, diagnostic, production, research, teaching)
- Nature of material (infectious, non-infectious)
- Dates of most recent information from laboratories
- Monitoring of the accuracy of reports
- Forms and sample letters to be sent to agencies/institutions can be found in Annex 2.

Laboratory inventory for materials that may contain wild poliovirus

The information obtained from laboratory inventories provides the data for a national inventory system. The purpose of the inventories is:

- to document location and type of wild poliovirus infectious and/or potentially infectious materials;
- to meet the country requirements for Regions to be certified as poliomyelitis-free; and
- to maintain a current list of laboratories for notification to initiate final containment procedures one year after the last wild poliovirus is detected globally.

Data to be submitted by each laboratory to its parent agency/institution should include documentation of search, type of material (infectious and potentially infectious), whether materials are contained, description and amount of each type of material, location in the laboratory where stored and documentation of BSL-2/polio storage conditions.

A flow chart for the Laboratory Inventory and forms to be completed and submitted to the laboratory’s parent agency/institution are included in Annex 3.
Annex 1

MATERIALS FOR NATIONAL LEVEL CONTAINMENT ACTIVITIES

Contents:

1. **Sample letter from National Authorities to Agencies/Institutions**: to be used as a model or sent directly to Agencies/Institutions selected by the National coordinator/group to be included in the National Plan for Containment.

2. **National Inventory Form**: to be compiled from data collected from all agencies/institutions and submitted to the WHO European Regional Office by **March 2002**. The forms along with all supporting documentation should be sent by mail, e-mail, or fax to:

   Dr George Oblapenko, Medical Officer
   Poliomyelitis Eradication
   WHO Regional Office for Europe
   8, Scherfigsvej
   2100 Copenhagen Ø
   Denmark
   Tel: 45 39 17 12 94
   Fax: 45 39 17 18 63
   E-mail: obl@who.dk
Sample letter from national authority to agencies/institutions with laboratories that might possess wild poliovirus or potentially infectious materials

Dear (organization/institution head):

At the request of the Director of the WHO European Region, I am writing to seek your assistance in laboratory containment of wild poliovirus infectious and potentially infectious materials. Your Agency/Institution is requested to:

- review the objectives of this activity as outlined in the enclosed materials;
- conduct a search of laboratories in your organization for wild poliovirus infectious and/or potentially infectious materials;
- establish an Agency/Institution Inventory of all laboratories that contain such materials.

In 1988, the World Health Assembly adopted the resolution calling for global eradication of poliomyelitis by the year 2000. Major regions of the world are now free of confirmed cases of poliomyelitis for years. Progress in European Region Member States is approaching the best expectations, pending further improvements in surveillance.

In few years, the only potential source of wild polioviruses, will be in the diagnostic, research, teaching, and vaccine production laboratories of the world. Unless the world laboratories fully prepare now for the time when poliomyelitis immunization stops, a chance reintroduction of poliovirus from one of these laboratories into the community could represent a public health threat of global proportions.

In Geneva in May 1999, the Fifty-second World Health Assembly reaffirmed WHO’s commitment towards the global eradication of poliomyelitis by the end of the year 2000, and urged poliomyelitis-free Member States to begin, in collaboration with WHO, the process leading to the laboratory containment of polioviruses.

Before regional certification can be approved, all member countries are required to search for and inventory all biomedical laboratories that might possess wild poliovirus infectious and/or potentially infectious materials. The search for laboratories not only includes those associated with traditional poliomyelitis laboratory functions but also includes laboratories in other sectors outside the Ministry of Health that could potentially have materials in storage collected during a time and in a place when wild poliovirus was circulating. The probability of a laboratory-associated poliovirus infection is small, but the consequences grow greater with time. When poliomyelitis immunization stops, a chance reintroduction of poliovirus from one of these laboratories into the community will represent a public health threat of global proportions.

The enclosed Guidelines for Implementation of Laboratory Containment of Wild Poliovirus for the European Region describe steps to be taken to prevent transmission of wild polioviruses from the laboratory to the community. A search of biological laboratories in each country and the establishment of National and Agency/Institutional Inventories are critical for implementing that Plan.

Please request all biomedical laboratories in your organization to carefully review their laboratories for infectious and potentially infectious materials according to Annex I found in the Guidelines. Each laboratory should provide documentation that they do not have, have appropriately disposed of, or have retained such materials. Any laboratory possessing such materials and wishing to retain these should be listed in the National Inventory and be certified as a BSL-2/polio level.

Please urge laboratories to dispose of all wild poliovirus infectious and potentially infectious materials no longer needed for current work. The enclosed Guidelines should be consulted for appropriate methods of destruction or for the addresses of the designated WHO/EURO interim Poliovirus Repositories.
Please keep this office informed of any change in the status of such materials that are in the possession of your institution, including reception/storage of new materials classified as wild poliovirus or potentially infectious materials.

One year after the last isolation of wild poliovirus worldwide, agencies/institutions listed in the National Inventory will be notified to instruct all laboratories to initiate procedures for maximum containment as described in the Action Plan for Laboratory Containment of Wild Polioviruses. This means destroying infectious and potentially infectious materials, upgrading facilities containing such materials to BSL-3/polio, or shipping materials to the designated wild poliovirus repository.

Please return the completed and signed laboratory inventory to me by … dates … .

Sincerely,

(National Authority)

Enclosures:
1. Action Plan for Laboratory Containment of Wild Polioviruses in the WHO European Region
2. Guidelines for Implementation of Laboratory Containment of Wild Poliovirus for the European Region
Official Form: National inventory of laboratories that have still stored wild poliovirus infectious and/or potentially infectious materials

(Report to be forwarded upon completion to the WHO Regional Office and National Committee for the Certification of the Eradication of Poliomyelitis)

Date: __________________________

National Authority: ____________________________________________________________________

Address:_____________________________________________________________________________

Phone number:___________________ Fax: _________________ E-mail: ________________________

Report prepared by: _____________________________________

NOTE: Completion of a National Inventories is a prerequisite for certification of the European Region as poliomyelitis-free. One year after detection of the last wild poliovirus, WHO will request countries to notify all agencies/institutions on the National Inventory to instruct laboratories to begin implementation of procedures for maximum laboratory containment of wild polioviruses as described in the Action Plan for Laboratory Containment of Wild Polioviruses.

This national authority has conducted a search of all agencies/organizations for laboratories with wild poliovirus infectious and potentially infectious materials as described in the Action Plan for Laboratory Containment of Wild Polioviruses.

We understand that one year after global detection of the last wild poliovirus, WHO will request countries to notify all agencies/institutions listed in the National Inventory to inform laboratories to begin implementation of procedures for laboratory containment of wild polioviruses, as described in the Action Plan for Laboratory Containment of Wild Polioviruses. This means destroying infectious and potentially infectious materials, upgrading facilities containing such materials to BSL-3/polio, or shipping materials to the designated wild poliovirus repository.

Number of agencies/organizations searched: _______________

Number of agencies/organizations that replied: _____________

I certify that the information on these pages is an accurate report of wild poliovirus infectious and potentially infectious material currently found in all laboratories under my jurisdiction. Any change in status will be reported on an annual basis as required by the WHO.

Signed:____________________________________ (representative of National Authority)

Date: _____________________________________
<table>
<thead>
<tr>
<th>Name and address of Agency/Institution (list all contacted including those declaring no materials)</th>
<th>Number of Control Laboratories</th>
<th>Number of Diagnostic Laboratories</th>
<th>Number of Production Laboratories</th>
<th>Number of Research Laboratories</th>
<th>Number of Teaching Laboratories</th>
<th>Date of most recent information</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Potentially infectious only</td>
<td>Infectious or both</td>
<td>Potentially infectious only</td>
<td>Infectious or both</td>
<td>Potentially infectious only</td>
<td>Infectious or both</td>
</tr>
</tbody>
</table>

National Inventory of Laboratories (According to type and possession of wild poliovirus infectious or potentially infectious materials)
Annex 2

MATERIALS FOR AGENCY/INSTITUTION CONTAINMENT ACTIVITIES

Contents:

1. Sample letter from Agencies /Institutions to laboratories
2. Agency/Institution inventory form
Sample memo from agency/institute head to laboratories requesting
inventory of wild poliovirus infectious or potentially infectious materials

To: Laboratory Director

From: Agency/Institute Director

The ___________ (government agency) has requested this ____________ (agency/institute) to search all biomedical laboratories under its jurisdiction that might possess wild poliovirus infectious and/or potentially infectious materials and to assist in carrying out a National Survey and in establishing a National Inventory of agencies/institutions that want to retain such materials.

In 1988 the World Health Assembly adopted the resolution calling for global eradication of poliomyelitis by the year 2000. In May 1999, the World Health Assembly reaffirmed WHO’s commitment towards the global eradication of poliomyelitis by the end of the year 2000, and urged poliomyelitis-free Members States to begin, in collaboration with WHO, the process leading to the laboratory containment of polioviruses.

In few years, the only potential source of wild polioviruses, will be in the diagnostic, research, teaching, and vaccine production laboratories of the world. Unless these laboratories fully prepare now for the time when poliomyelitis immunization stops, a chance reintroduction of poliovirus from one of these laboratories into the community could represent a public health treat of global proportions.

The enclosed Guidelines for Implementation of Laboratory Containment of Wild Polioviruses for the European Region describes steps to be taken to prevent transmission of wild polioviruses from the laboratory to the community. A search of biomedical laboratories in each country and the establishment of National and Agency/Institutional Inventory are major steps toward implementing that Plan.

The enclosed Guidelines give definitions and examples of materials included in the national survey and to be listed in the laboratory inventory. Laboratories are urged to dispose of all such materials for which there is no longer a need. Guidelines for proper disposal of such materials are enclosed (attachment V)

I am asking that you carefully review the enclosed documentation/guidelines in order to fully document that your laboratory does not possess such materials, has properly disposed of such materials, or has accurately identified all such materials in its possession and will adhere to the biosafety requirements.

Laboratories wishing to retain or dispose of wild polioviruses or potentially infectious materials should consult the Guidelines to insure proper methods are used. Laboratories retaining wild polioviruses or potentially infectious materials must do so for the moment under biosafety level-2/polio (BSL-2) conditions, with secure storage and a current inventory at all times. These laboratories will be requested to upgrade the biosafety levels after certification of eradication of poliomyelitis.

Laboratories containing such materials will be listed in the National Inventory of Agencies/Institutions That Possess Wild Polioviruses and Potentially Infectious Materials, which is maintained in the ___________ (government agency). The Office _________ (of this institution) should be kept informed of any changes in the inventory.

Please return the completed form to the Office___________(of this institution) by ________(date).

Sincerely,

Director of Institution/Organizations

Enclosure: Guidelines for Implementation of Laboratory Containment of Wild Polioviruses
Official Form: Agency/institution inventory of laboratories that contain wild poliovirus
infectious and/or potentially infectious materials
(Report to be forwarded upon completion to National Authority)

Date: __________________________

Agency/Institution: ________________________________________________________________

Address: _______________________________________________________________________

Phone number: __________________ Fax: _________________ E-mail: ______________________

Report prepared by: _______________________________________

_______________________________________________________________________________

This agency/institution has conducted a search of all laboratories under its jurisdiction for wild
poliovirus infectious and potentially infectious materials as described in the Action Plan for
Laboratory containment of Wild Polioviruses.
Number of laboratories searched: __________
Number of completed inventories returned: __________

(Please check (✓) one of the following.)

☐ This agency/institution has no laboratories that possess wild poliovirus infectious or potentially
infectious materials.

☐ The following laboratories possess wild poliovirus infectious and/or potentially infectious materials
(refer to Table 1 next page).
### Table 1. Agency/Institution Inventory

<table>
<thead>
<tr>
<th>Name and address of laboratory</th>
<th>Type of laboratory*</th>
<th>Does laboratory meet BSL-2/polio requirements?**</th>
<th>Listing of materials</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Potentially infectious only</td>
</tr>
<tr>
<td>1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td></td>
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<td>3</td>
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<td>7</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>8</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* Select one of the following that best describes the activities of the laboratory: **C** = control, **D** = diagnostic, **P** = production, **R** = research, or **T** = teaching.

** Refer to attachment detailing BSL-2/polio requirements.

I understand that one year after global detection of the last wild poliovirus, WHO will request countries to notify all agencies/institutions on the **National Inventory** to instruct laboratories to implement procedures for laboratory containment of wild polioviruses as described in the **Action Plan for Laboratory Containment of Wild Polioviruses**. This means destroying infectious and potentially infectious materials, upgrading facilities containing such materials to BSL-3/polio, or shipping to the designated wild poliovirus repository.

I certify that the above is an accurate report of wild poliovirus infectious and potentially infectious materials currently being contained by all laboratories under my jurisdiction. Any change in status will be reported on an annual basis as required by the National Registry.

Signed: ___________________________________________ __________________________

Director of Agency/Institution Date
Annex 3

MATERIALS FOR LABORATORY CONTAINMENT ACTIVITIES

Contents:

1. Flow chart for laboratory search of all laboratory spaces (including refrigerators/freezers) and materials

2. Laboratory inventory form: to record all infectious or potentially infectious materials retained by the laboratory

3. List of countries and dates of last poliovirus endemicity

4. Biosafety level 2 – polio requirements (BSL-2/polio)

5. Methods for disposal of poliovirus infectious or potentially infectious materials
The full collaboration of laboratory workers and supervisors at all levels is required to assure that inadvertent transmission of wild poliovirus from the laboratory to the community does not occur. This is a serious responsibility.

Biomedical laboratories are requested to carefully search all storage records and facilities to determine the presence or document the absence of wild poliovirus infectious and/or potentially infectious materials. Materials searched for are grouped into the following 2 categories:

**Infectious:** Any material (clinical, animal, research, environmental) known to contain wild poliovirus

- Wild poliovirus stocks (reference strains, isolates, and proficiency test panels) or research materials containing capsid sequences derived from wild polioviruses
- Any clinical material (throat, faecal or autopsy specimens) from confirmed or suspected poliomyelitis cases
- Animals infected with wild polioviruses
- Stored specimens from experimental animals infected with wild polioviruses

**Potentially infectious**

- Throat or faecal specimens **collected for any purpose** at a time or in a region where poliomyelitis was endemic or epidemic
- Environmental (water and sewage) specimens **collected for any purpose** at a time or in a region where poliomyelitis was endemic or epidemic
- Untyped enterovirus-like and undifferentiated poliovirus isolates

Non-poliovirus laboratories must take particular care to identify potentially infectious materials that may have been collected at a time or in a geographic area where poliomyelitis was endemic or epidemic. Please consult the accompanying list of countries and dates of last endemicity. Questions pertaining to specific materials, dates, or places should be referred to: Helpline/desk/Web site (or National Reference Laboratory for Polio Eradication) to answer specific questions.

Routinely collected serum specimens and cerebrospinal fluids are not likely to contain sufficient levels (if any) of poliovirus to cause infection and are not considered potentially infectious (unless obtained from known or suspected cases of poliomyelitis).

**Parent agency**

Any wild poliovirus infectious or potentially infectious materials that are no longer critical to the laboratory mission should be destroyed by autoclaving or as described in Attachment V: *Methods for Disposal of Poliovirus Infectious or Potentially Infectious Materials.*

The attached Laboratory Inventory form should be completed in full, using duplicate pages as necessary, and submitted to its parent agency/institution on or before the requested date. Submissions should also include written documentation of how and what facilities were searched.
The inventory of laboratories that keep strains of wild poliovirus should be kept current at all times. Any changes in numbers or locations of wild poliovirus infectious or potentially infectious materials should be recorded as they occur. The laboratory should notify its parent Agency/Institution of any changes in wild poliovirus status.

All poliovirus infectious or potentially infectious materials retained by the laboratory should be maintained under BSL-2/polio conditions (please refer to Attachment IV: Biosafety Level (BSL-2/polio requirements)). One year after detection of the last wild poliovirus worldwide, all laboratories on the National Inventory will be requested to destroy, transfer, or place such materials under high containment conditions.

Thank you for your help in making the world safe from poliomyelitis.

Attachments:

I. Flow chart for inventory of wild poliovirus infectious and/or potentially infectious materials
II. Official laboratory inventory form
III. List of countries and dates of last known wild poliovirus endemicity
IV. Biosafety level (BSL)-2/polio requirement
V. Methods for disposal of poliovirus infectious or potentially infectious materials
Attachment I: Flow chart for inventory of wild poliovirus infectious and/or potentially infectious materials

How many freezers/refrigerators are located in this laboratory? (Record # on form)

Has material been stored under conditions known to favour virus survival?
Excluded are specimens stored without refrigeration for 90 days or more, refrigerated for one year or more, heat inactivated, treated with a disinfectant known to inactivate polioviruses, or tested and found negative for the presence of enteroviruses.

Yes

Infectious

Is the material a wild poliovirus stock (reference strain, isolate, or proficiency test panel) or research product containing capsid sequences derived from wild polioviruses?

Is the material a clinical specimen (throat, faecal or autopsy specimen) from a confirmed or suspected case of polio?

Is the material a stored specimen from an experimental animal infected with wild poliovirus?

Is the material untyped enterovirus-like or undifferentiated poliovirus cell cultures isolate?

Is the material a throat or fecal specimen from a study or field survey?

Not potentially infectious or infectious

No

Potentially infectious

Not an infectious material

Was the material collected at a time or in a geographical area where polio was endemic? (consult attached list of countries)

Yes

Review all wild poliovirus infectious and potentially infectious materials
Are they needed by the laboratory?

No

Destroy and document or ship wild poliovirus stocks to designated repository

Complete inventory form and return to head of agency/institution

Maintain BSL-2/polio conditions
Implement on-going inventory of wild poliovirus stocks and potentially infectious materials

Yes
Attachment II: Official laboratory inventory of wild poliovirus infectious and/or potentially infectious materials

(Duplicate report upon completion to be forwarded to requesting agency/institution)

Date:_________________________________

Laboratory:____________________________ Type of laboratory
(please check (√) the appropriate boxes):
❑ Control       ❑ Diagnostic
❑ Production   ❑ Research
❑ Teaching     ❑ Other ________________

Institution: __________________________________________________________________________

Address:____________________________________________________________________________

Phone number:______________ Fax: ________________ E-mail: _______________

Report prepared by: ________________________________________

(Please check (√) one or more of the following.)

❑ This laboratory has carefully searched its records and storage facilities and has not found or has not destroyed any wild poliovirus infectious or potentially infectious materials.
  (Sign and return to requesting agency/institution).

❑ This laboratory has carefully searched its records and storage facilities and has destroyed all wild poliovirus infectious or potentially infectious materials. (Please complete laboratory inventory form, next page and see attachment V, Methods for disposal of poliovirus infectious or potentially infectious materials).

❑ This laboratory wishes to retain the following wild poliovirus and/or infectious or potentially infectious materials. (Please complete laboratory inventory form, next page.)
Laboratory inventory form for poliovirus containment

Name of laboratory: _____________________ Investigator name: _____________________ Date: _____________

# of refrigerators/freezers in lab: __________ # of refrigerators/freezers searched: __________

**MATERIALS SECTION**

<table>
<thead>
<tr>
<th>Type of material</th>
<th>Description of material and amount</th>
<th>Date/place of collection</th>
<th>Location and conditions of storage</th>
<th>Storage history</th>
<th>Future of material*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reference strains</td>
<td></td>
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<td></td>
</tr>
<tr>
<td>Isolates</td>
<td></td>
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<tr>
<td>Proficiency test panels</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Research materials with capsid sequences from wild polioviruses</td>
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<td></td>
</tr>
<tr>
<td>Throat specimens</td>
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<td></td>
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<tr>
<td>Faecal specimens</td>
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<tr>
<td>Autopsy specimens</td>
<td></td>
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<td></td>
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<tr>
<td>Faecal specimens</td>
<td></td>
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<td></td>
</tr>
<tr>
<td>Throat specimens</td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Non-human primates infected with wild polioviruses</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Transgenic mice infected with wild polioviruses</td>
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<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Stored specimens from experimental animals</td>
<td></td>
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<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Undifferentiated Poliovirus isolates</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Untyped enterovirus-like isolates</td>
<td></td>
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</tr>
<tr>
<td>Water samples</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sewage samples</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Please indicate whether material will be **D** = destroyed; **S** = shipped; **M** = maintained under BSL-2/polio.
We certify that the above is an accurate record of wild poliovirus infectious and/or potentially infectious materials currently in possession of this laboratory and maintained under BSL-2/polio conditions, including secured storage area with limited access.

This laboratory has searched all relevant storage areas for wild poliovirus infectious and potentially infectious materials as described in the Action Plan for Laboratory Containment of Wild Polioviruses. We understand that one year after global detection of the last wild poliovirus, WHO will request countries to notify all agencies/institutions listed in the National Inventory. These agencies/institutions will be instructed to inform laboratories to begin implementation of procedures for laboratory containment of wild polioviruses, as described in the Action Plan for Laboratory Containment of Wild Polioviruses. This means destroying infectious and potentially infectious materials, upgrading facilities containing such materials to BSL-3/polio, or shipping materials to the designated wild poliovirus repository.

Signed:____________________________________ ____________________________
______________________________
Director of Laboratory Date

___________________________________ ____________________________
______________________________
Director of Agency/Institute Date
<table>
<thead>
<tr>
<th>Country</th>
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Attachment IV: Biosafety level (BSL)-2 / polio requirements

1. Good microbiological techniques are practiced.
   - Specimens are handled safely
   - Pipettes and pipetting aids are used safely
   - Ingestion of infectious materials is avoided
   - Centrifuges are used safely
   - Refrigerators are maintained and used safely
   - Ampoules containing infectious materials are opened safely
   - Appropriate disinfection and sterilisation are carried out
   - Hands are washed between procedures and prior to leaving laboratory
   - Laboratory gowns are worn for work in laboratory and not worn out of the laboratory
   - Storage of food or drink in the laboratory or any storage receptacle containing infectious materials is prohibited
   - Eating, drinking, or smoking in the laboratory is prohibited
   - No mouth pipetting is permitted
   - Dispersal of infectious materials is avoided
   - Separation of serum is carried out safely
   - Tissue grinders are used safely
   - Infectious materials are stored safely
   - Homogenizers, shakers and sonicators are used safely
   - Contact of infectious materials with skin and eyes is avoided
   - Precautions are taken with blood and other bodily fluids
   - Specimens and infectious materials are shipped safely

2. Facility meets standards for basic BSL-2 laboratory.
   - Walls, ceilings and floors are easily cleanable.
   - Illumination is adequate for all activities.
   - Autoclaves are available to sterilize contained material.
   - Access to laboratories is restricted.
   - Storage space is adequate to hold supplies for immediate use.
   - Hand washing basins, with running water, if possible, are provided in each laboratory room, preferably near the door.
   - An autoclave (or suitable pressure cooker) is available in the same building as the laboratory.
   - Facilities for storing outer garments and personal items for eating and drinking are provided outside the working areas.
   - A good-quality and dependable water supply is available. There are no cross-connections between sources of laboratory and drinking-water supplies.
   - A standby generator is desirable for the support of essential equipment such as incubators, biological safety cabinets, freezers, and the like.
   - Pipetting aids are available to replace mouth pipetting.
   - Biological safety cabinets are available for:
     - Procedures with high potential for producing aerosols, including centrifugation, grinding, blending, vigorous shaking or mixing, sonic disruption, and opening of containers of infectious materials whose internal pressure may be different from the ambient pressure, handling high concentrations or large volumes of infectious materials.
     - Centrifuges with sealed safety cups or brackets are available for centrifuging high concentrations or large volumes of infectious materials in the open laboratory. These cups or brackets must be loaded and unloaded in a biological safety cabinet.
     - Screw-capped tubes and bottles are available to hold positive specimens and cultures.

3. Persons entering the laboratory have been fully immunized against poliomyelitis.

4. Use of wild polioviruses is discontinued where attenuated vaccine polioviruses, inactivated antigens, or non-polio enteroviruses may serve the same purposes, for example, as challenge viruses in neutralizing antibody tests.

5. All poliovirus stocks and potentially infectious materials are disposed of when there are no programmatic or research needs for retention.

6. An internal control system is implemented for all wild polioviruses contained in the laboratory (current inventory, good record keeping).

7. Wild polioviruses are stored in separate, secure areas with limited access.

8. Only viruses that are readily identifiable by molecular methods are used if wild virus reference strains or working stocks are required.

9. Appropriate sterilization and/or incineration is used for disposing of wild polioviruses, infectious materials and potentially infectious materials.
Attachment V: Methods for disposal of poliovirus infectious or potentially infectious materials

Sterilization (use of autoclaves)

Moist steam under pressure is the most effective method of sterilization of laboratory materials.

- All cultures and contaminated materials should normally be autoclaved in leak proof containers, e.g., autoclavable, color-coded plastic bags, before disposal.
- Plastic bags should be opened so that steam will penetrate to their contents.
- After being autoclaved, the materials may be placed in transfer containers for transport to the incinerator or other point of disposal.

Incineration

- Incineration is the method of choice for final disposal of contaminated waste, including carcasses of laboratory animals, preferably after autoclaving. Incineration of infectious materials is an alternative to autoclaving only if:
  - the incinerator and transport to the incinerator is under laboratory control;
  - the incinerator is provided with an efficient means of temperature control and a secondary burning chamber.
- Materials for incineration, even if they have first been autoclaved, should be transported to the incinerator in bags, preferably plastic.
- Incinerator attendants should implement proper procedures for loading and temperature control.

Final disposal

The disposal of laboratory and medical waste is subject to various national regulations. In general, ash from incinerators may be treated in the same way as normal domestic waste and removed by local authorities. Autoclaved waste may be disposed of by off-site incineration or in licensed landfill sites.

Source: World Health Organization