EXPERT COMMITTEE ON BIOLOGICAL STANDARDIZATION
Geneva, 17 to 21 February 2003

GUIDELINES FOR THE SAFE PRODUCTION AND QUALITY CONTROL OF IPV MANUFACTURED FROM WILD POLIOVIRUSES

BIOSAFETY LEVEL 3/POLO

NOTE: This document has been prepared for the purpose of inviting comments and suggestions on the proposals contained therein, and for the preparation of the materials to be considered by the Expert Committee on Biological Standardization. The text in its present form does not necessarily represent an agreed formulation of the Expert Committee. Comments proposing modifications to this text MUST be received by 31 January 2003 and should be addressed to the World Health Organization, 1211 Geneva 27, Switzerland, attention: Quality Assurance and Safety of Biologicals (QSB).

Addendum to WHO Recommendations for the Production and Quality Control of Poliomyelitis Vaccine (Inactivated).

The WHO Recommendations for Production and Quality Control of Poliomyelitis Vaccine (Inactivated) were last revised in 2000. At that time it was envisaged that production and quality control of inactivated poliomyelitis vaccine (IPV) manufactured from wild poliovirus strains should, in the near future, comply with increased laboratory biosafety conditions. This was because of the context of an increasingly polio-free world and the need for effective containment of wild poliovirus strains as a pre-condition of global certification of polio eradication.
These guidelines specify steps to minimise the risk of reintroducing wild poliovirus from a vaccine manufacturing facility into the community after global certification of polio eradication. Each of the following sections constitutes guidance for national control authorities and for the manufacturers of inactivated poliomyelitis vaccine. If a national control authority so desires, these Guidelines may be adopted as definitive national requirements, or modifications may be justified and made by a national control authority. It is recommended that modifications to these Guidelines be made only on condition that the modifications ensure that the risks of reintroducing wild poliovirus to the community are no greater than as outlined in the Guidelines set out below.

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1. INTRODUCTION

In May 1999, the World Health Assembly reaffirmed the commitment of the World Health Organization (WHO) to eradicate poliomyelitis and urged all member states to begin the process leading to the containment of stores of wild poliovirus. In December 1999, after consultation with scientists, ministries of health, and vaccine manufacturers worldwide, WHO published the *WHO global action plan for laboratory containment of wild polioviruses*. A second edition of the Global Action Plan published in 2003 replaces the first. The purpose of the plan was to identify steps that all countries should take to minimize the risk of reintroducing wild poliovirus from a laboratory or vaccine production facility into the community after polio eradication. The plan calls for materials infected or potentially infected with poliovirus to be handled and stored under biosafety conditions appropriate to their risk, with implementation of these containment levels a requirement for the certification of eradication. For facilities such as IPV manufacturers, where infectious materials come into contact with permissive cells or animals, high containment (Biosafety Level 3/polio) measures are required.

Biosafety Level-3/polio requires the primary and secondary containment of wild poliovirus infectious materials, with provisions governing air, water, and materials entering and leaving the facility, specific requirements for personal protective clothing, laboratory design, the use of laboratory equipment, and medical surveillance of laboratory staff. Additionally it requires vaccination of all staff, appropriate training for biosafety procedures, and validation and documentation of the physical and operational requirements. Implementation of such containment conditions within IPV production and quality control testing facilities must also take into account the large quantities and concentrations of live vaccine that are produced, the industrial scale of facilities, as well as the existing rules and regulations governing the manufacture and testing of medicinal products, commonly known as Good Manufacturing Practices (GMP).

Both biosafety and GMP share the control of contamination as one of their major concerns. Where GMP prioritizes the safety of the patient being treated with the medicinal product, biosafety is primarily concerned with the protection of the personnel and the surrounding environment. These concepts are not mutually exclusive, and a considerable area of overlap exists between the two. It may not always be possible for facilities and procedures to meet the ideal situation as seen from both GMP and biosafety perspectives, and some degree of flexibility may be required to satisfy both objectives. A number of the internationally harmonized GMP guidelines, such as those of the WHO or the European Union, have introduced specialized GMP requirements for biological products. The requirements for IPV producers in the post-eradication era should be viewed as a special addendum to be read with these guidelines, where a licensed medicinal product must be manufactured and tested within a large-scale containment facility in the exceptional case where, through the combined efforts of many partners, the disease for which the vaccine has been used has been eliminated globally.
The scope of this document addresses the period after the interruption of wild poliovirus circulation. At that time, natural immunity due to contact with wild poliovirus will start to decline, but immunization coverage is anticipated to remain adequate to protect both individuals and communities. The containment measures outlined in this document will remain in force as long as universal polio immunization continues. If polio immunization is discontinued in some or all countries after global certification, containment requirements for wild as well as OPV viruses will need to be reconsidered and may become more stringent, consistent with the consequences of inadvertent transmission of wild poliovirus from the laboratory or vaccine production facility to an increasingly non-immune community. These guidelines have been produced in the full recognition that adequate global capacity for IPV production must be maintained while effective biosafety measures are in place, and this will remain the shared responsibility of the global IPV manufacturers, their national oversight authorities for medicines and the environment, and the partner organizations active in the global eradication of polio.

2. BIOSAFETY IMPLEMENTATION WITHIN A VACCINE PRODUCTION FACILITY

A breach of containment of poliovirus used in a vaccine production or testing facility can theoretically occur through contaminated clothing, liquid or air effluents, or improper virus disposal. Transmission from the laboratory or vaccine production facility to the community is most likely to result from either equipment failure or human error. Of greatest concern is the inadvertent transmission of poliovirus to the community through an infected laboratory or vaccine production worker. There is also a possibility of an unexpected emergency that could lead to release of infectious materials into the community. The provisions in these guidelines seek to minimize the risk of these occurrences.

2.1 Biosafety Level-3/polio requires that the institution employ a Biosafety Officer who is knowledgeable in large-scale viral production and containment, but is independent of production in his or her reporting structure. The Biosafety Officer is responsible for the independent oversight of the implementation of the biosafety practices, policies, and emergency procedures in place within the company or organization. A Biosafety Officer is needed in addition to a Qualified Person who, in some countries, has overall responsibility for a medicinal product.

2.2 There should also be a Biosafety Committee comprising representatives of viral production and quality control that functions to review the biosafety status within the company and to coordinate preventative and corrective measures. It must include the institutional Biosafety Officer as a member.

2.3 A detailed and comprehensive risk analysis should be conducted to define
possible contamination sources to personnel or the environment that may arise from the production or testing of live poliovirus within the establishment. For each procedure or system, this analysis should take into account the concentration and stability of the virus at the site, the potential for inhalation, ingestion, or injection that could result from accidents, and the potential results of a major or minor system failure. The procedural and technical measures to be taken to reduce the risk to workers and the environment should be considered as part of this analysis. The results of this risk analysis should be documented.

2.4 A comprehensive Biosafety Manual must be created and implemented that fully describes the biosafety aspects of the production process and quality control activities and defines such items as emergency procedures, waste disposal, and the requirements for safety practices and procedures as identified in a risk analysis. It should be made available to all staff of the production and quality control units, with at least one copy present in the containment area(s). The manual should be reviewed and updated annually.

2.5 Comprehensive guidelines outlining the response to biosafety emergencies and accidents should be prepared and made available to key personnel for information and for coordination with emergency response units. These guidelines should be reviewed and updated annually.

2.6 The implementation of Biosafety Level 3/polio status in the production and testing facilities should be verified through an independent assessment. Current national requirements concerning verification mechanisms should be complied with.

3. PERSONNEL

3.1 The manufacturing establishment and its personnel shall be under the authority of a qualified person who has been trained in the techniques used in manufacturing biological substances and who possesses the scientific knowledge upon which the manufacture of these products is based.

3.2 Personnel required to work in the polio virus containment area(s) should be selected with care to ensure that they may be relied upon to observe the appropriate codes of practice and are not subject to any disease or condition that could compromise the integrity of the product or the safe containment of the poliovirus strains with which they work.

3.3 Health examinations of personnel should be required before employment and periodically (at least annually) thereafter. Any changes in health status that could adversely affect the quality of the product or the containment procedures should preclude the person concerned from working in production
or testing of live poliovirus. Immune compromised individuals should not be permitted to work in the containment area. Attention should be paid that adequate precautions have been taken to minimize the risk of transmission of poliovirus from personnel to their immediate family members.

3.4 All personnel (including those concerned with cleaning, maintenance or quality assurance) employed in areas where live poliovirus is manufactured or tested should receive additional training specific to their work with poliovirus. This should include relevant information and training in hygiene and microbiology as it relates to IPV and poliovirus, as well as basic principles of BSL-3/polio containment procedures. Personnel coming in direct contact with poliovirus-containing materials in the production and quality control area, including technicians and support personnel, must demonstrate proficiency in standard microbiological practices as outlined in the WHO Biosafety Manual as appropriate to the production and testing of IPV. Records of this training should be maintained and periodic assessments of the effectiveness of training programs conducted.

3.5 Personnel engaged in IPV production and testing and all visitors to the production and testing facilities, including regulatory authorities, civil inspectors, or emergency personnel should be vaccinated with poliomyelitis vaccines, and adequate blood levels for circulating neutralizing antibody titres against all three serotypes should have been verified. Test results should be no older than two years. Titre results on staff should be kept indefinitely. To insure adequate mucosal immunity and reduce the likelihood of faecal shedding, it is strongly advised to immunize personnel with oral polio vaccine (OPV) alone or in addition to inactivated polio vaccine (IPV). In cases where OPV is not used for routine immunization in the country of manufacture, it may be necessary to employ special arrangements for procuring vaccine for persons at high risk of infection.

3.6 After any major spill or accident where there is reason to believe that the personnel may have been infected with poliovirus, national authorities should be notified and the appropriate emergency containment and monitoring procedures initiated. Diagnostic examinations of personnel should be carried out to determine whether they are shedding poliovirus in their faeces. If poliovirus is being shed in faeces, appropriate measures as detailed in the facility guidelines for responding to biosafety emergencies and incidents shall be employed to avoid transmission of poliovirus to other persons or to the public sewage system.

4. PREMISES AND EQUIPMENT

Premises should be designed in such a way as to control both the risk to the product and to the environment. This is accomplished by establishing a primary containment barrier, through appropriate safety equipment to protect the immediate workspace, and
enclosing this system within a secondary containment barrier that protects the environment external to it from accidental exposure to infectious materials. These systems must provide adequate safeguards to protect the product against contamination with extraneous agents depending on the environmental cleanliness level required by the operation.

General requirements

4.1 Live poliovirus and materials with a potential to contain live poliovirus should be handled in contained areas. Contaminated materials, including equipment for repair or maintenance, should be decontaminated by a validated method prior to removal from the containment area.

4.2 Whenever possible, polio production facilities and quality control (QC) facilities should be in dedicated buildings. If they are located in multipurpose buildings, the polio production facility must have separate access and exits for personnel and materials, and dedicated air and biological waste handling systems must be provided. Polio QC laboratories in multi-purpose buildings should be equipped with air handling and waste disposal systems that preclude the contamination of other areas with poliovirus infectious material.

4.3 Production for other organisms on a campaign basis may be acceptable within the poliovirus facility provided that the facilities are disinfected following poliovirus production using a validated area fumigation procedure.

4.4 Laboratories and production areas should be marked with approved biohazard signs. Information should be posted in prominent locations at the entry to the BSL-3/polio facility that poliovirus is contained in the area, that immunization against poliomyelitis is required for entry, and that only personnel authorized to work with poliovirus are permitted to enter. The name(s) and contact information of persons to contact in the event of an emergency should be presented and kept up to date at all times.

4.5 All exits must be marked. Emergency exit doors from the polio facility must be alarmed and their use treated as a breach of containment.

4.6 Protective laboratory or production clothing such as solid-front or wrap-around gowns, scrub suits, or coveralls with head and shoe covers should be worn at all times by operators while in the containment facility. Eye covers or full-face masks should be required when there is a potential for generating aerosols. Respirators should be used when conducting procedures with a high probability for aerosol generation. Disposable gloves should be worn when handling infectious materials or contaminated equipment. This protective clothing is not to be worn outside the facility. Clothing should be decontaminated before being laundered or disposed.
Security access control

4.7 Access to the plant or institution grounds should be monitored and appropriate security measures should be put in place to avoid the entry of unauthorized persons or intruders at any time.

4.8 Access to the polio vaccine production facility or polio vaccine quality control testing laboratories should be strictly limited to personnel with authorization to enter the specific area. Entrance into the facility should be monitored at all times. An example of a currently accepted procedure would be a magnetic badge controller on self-closing locked entry doors.

Primary containment barriers

4.9 Biological safety cabinets should be provided and used within the production and quality control areas where live virus or infected cell cultures are handled or manipulated and where such activities cannot be carried out in closed transfer systems. Positive pressure laminar flow hoods shall not be substituted for negative pressure biological safety cabinets when handling potentially infectious material.

4.10 Biological safety cabinets must be constructed and manufactured in accordance with national regulations or standards, such as BSI, DIN or NSF. They must be tested and certified at least annually to meet those standards. Cabinets with design modifications to meet the constraints of large-scale operations but providing equivalent containment levels may be utilized if approved by the responsible national authorities.

4.11 When exhaust air from biological safety cabinets is to be discharged through the building exhaust air system, the air handling system must be designed in such a way as to not disturb the air balance of the cabinet or the room.

4.12 Bio-reactors should be designed wherever possible as closed systems with entry and exit ports that do not require open manipulation of viable poliovirus in the production room, e.g. using steam-through valves or sterile tube welders. Bio-reactors should have the capability of being sterilized while loaded to their maximum capacity. Bioreactor air vents should be provided with high efficiency particulate air (HEPA) filters or an equivalent system to sterilize exhaust gasses.

Secondary containment barriers

4.13 Containment areas should be separated from access corridors by separate airlocks for personnel and materials. Airlocks should consist of one or more
closed chambers and be equipped with interlocking doors or an equivalent system to ensure that both doors cannot be opened simultaneously. Personnel and material airlocks with doors leading to the containment area should be provided with a ducted ventilation system that exhausts air through a HEPA filter. Adequate time should be allowed for the air handling system to flush out contaminants that have entered the airlock from the containment area before opening the door leading to the exterior. When possible, separate airlocks for the entrance and exit of personnel should be provided.

4.14 Airflow patterns should not present a contamination risk. Care should be taken to ensure that airflows do not distribute poliovirus from a zone of higher contamination risk to a zone of lower contamination risk. A pressure differential of at least 10-15 Pa should be maintained at all times between zones.

4.15 An air handling system should maintain a negative pressure (inward directional airflows) in areas where live poliovirus is handled or there is a potential for contamination. Air filtration can be accomplished by the installation of HEPA filters providing a filter efficiency of 99.97% or greater removal of 0.3-micrometer particles. Air from areas where live poliovirus is handled or there is a potential for contamination should be extracted through HEPA filters at the point of air removal from the chamber or airtight ducts. Although not normally recirculated, air may be recirculated to the same area provided it is HEPA filtered before reuse. A proper system for maintenance and testing of HEPA filters must be in place. Heat exchangers may be utilized to recover warmth from HEPA filtered exhaust air.

4.16 HEPA filters should be installed into the air handling systems in such a manner as to allow gaseous decontamination of the filters before removal or testing by accepted aerosolised challenge methods. Such filters must be tested and certified upon installation and at least annually thereafter.

4.17 Indicators of pressure differences should be fitted where these differences contribute to containment. Pressure difference readings for rooms or across HEPA filters should be monitored and recorded regularly or otherwise documented.

4.18 A warning system consisting of an audible or visual signal that can be readily perceived by personnel in the containment facility should be provided to indicate any failure in the air handling system.

4.19 The supply and exhaust air must be interlocked to prevent the positive pressurization of the containment area in the event of a failure of the system.
4.20 Exhaust air should provide sufficient air changes in both the QC and production areas to reach an appropriate environmental cleanliness level. At least 10 air changes per hour should be maintained.

4.21 Containment premises should be easily disinfected and should have the following characteristics:

a) There should be no windows that can be opened or any direct venting to the outside. Windows must be constructed of break resistant safety glass with strength characteristics conforming to the purpose for which they are used;

b) Passageways for pipes, tubes, and ducts passing through the wall between the containment area and surrounding areas should be completely sealed with materials resistant to contaminants and capable of withstanding disinfectants;

c) Floor drains where installed must be capped, fitted with liquid tight gaskets, or connected to a waste effluent decontamination system to prevent inadvertent release to the sanitary drain;

d) Wherever possible provisions should be made to contain liquids leaking from bio-reactors or tanks (including waste tanks) by means of floor dams or ramps that enclose a volume equal to the maximum fluid contained in the vessels plus the disinfectant required for inactivation;

e) All liquid and gas services to the containment area must be protected from back flow. Vacuum lines should be protected with liquid disinfectant traps and HEPA filters or 0.2μm hydrophobic membrane filters, or their equivalent.

4.22 If circulating water with open taps is used within the containment area, a spill or contamination at the point of use should not result in a breach in containment via the water system. Water loops should be maintained at 80°C or greater, and dead legs should be avoided. Heat exchangers may be used to cool water at the point of use provided that water from the exchanger is not returned to the loop leading outside of the containment area. If there is an accidental release of poliovirus in the areas served by the water loop, or if the circulating temperature of the water system drops below its set point to an extent identified by the risk assessment, an alarm should sound and the system temperature should be raised to a temperature and time period validated to kill poliovirus before taps can be opened outside or inside the containment area.

4.23 A communication system consistent with the facility containment conditions should be maintained between the support or administrative area and the containment area and shall be kept in working order at all times.
4.24 Emergency lighting and power to the containment area and critical containment devices (e.g. biological safety cabinets and air handling systems) should be available and automatically activated in the case of a power failure.

**Sterilization and waste disposal systems**

4.25 Decontamination of solid, liquid, and gaseous wastes should take place within the containment area. Should any wastes have to be transported out of the facility prior to decontamination and disposal, they must not be transported through public areas and must be packaged, labelled, and transported in accordance with applicable regulations.

4.26 The production unit must be provided with one or more interlocking, double door pass-through autoclaves that is validated at least annually. Liquid effluent from the sterilization chamber of the autoclave should go to the building liquid effluent sterilization system. Other means of sterilization of materials, such as fumigation with formaldehyde, gaseous peracetic acid, or vapour phase hydrogen peroxide may be used if they are fully validated for efficacy.

4.27 Liquid effluents from equipment, showers, and sinks within the containment area must be sterilized by autoclaving or discharge to a liquid effluent decontamination system. Such a system must be fully validated to ensure efficacy and be contained in an area compatible with the requirements of BSL-3/polio. The effluent tanks must be situated in an area with floor dams that will contain the full tank volume. If decontamination is to take place within the floor dam, the volume must be sufficient to add disinfectant to fully inactivate its contents.

4.28 Viral seeds must be stored within a BSL 3/polio containment area, or if in a separate location, under BSL 3/polio conditions in leakproof primary containment containers. Secondary containers for transfer of viral seeds from the storage area to the production area should be leak-proof and unbreakable.

4.29 The areas for the storage of viral seed stock must be dedicated and fully secured against entry by non authorised personnel.

4.30 The viral seed stock must be inventoried. Addition or removal of material must be conducted by authorized personnel following the approval of two authorized signatories on record or the electronic equivalent of this approval. Records of additions or removal of viral seed must be securely stored.

4.31 The viral seed storage area must be equipped with back-up emergency power source and recording and alarm systems to monitor freezers.
5. DOCUMENTATION AND VALIDATION

5.1 Detailed records of operating parameters for the containment facility should be produced and maintained for conducting an assessment of the facility performance.

5.2 All spills or accidental release of infected materials and their response should be properly investigated and documented. These investigations should be used to review and revise the facility and applicable operating procedures as required.

5.3 The production facility must be designed and constructed in such a manner as to allow for full validation of containment processes. It is the responsibility of the institution to ensure that these facilities meet acceptable standards that will ensure containment of poliovirus as well as the protection of the staff and the environment. These tests should be carried out at the completion of construction or renovation. Annual verification that the facility continues to meet the containment conditions should be performed. Records of the annual verification should be maintained throughout the lifetime of the IPV production facility and for at least 5 years after the facility stops production. At the minimum the following containment features should be assessed:

a) integrity of containment perimeter, including penetrations through floors, walls, and ceilings;

b) air tightness of supply and exhaust ductwork between incoming and first outgoing HEPA filter ducting in the air handling systems. The duct should be considered to be part of the room up to the point of the disinfecting filter or incinerator;

c) integrity of all HEPA filters and high efficiency filters and filter housings;

d) directional inward air flow from non-contained areas to containment areas;

e) biological safety cabinets and primary containment devices;

f) autoclaves, include cold spot and standard load testing using biological indicators or by means of physical validation;

g) waste effluent systems and holding tanks;

h) liquid back flow prevention devices;

i) alarm systems for air system failures, room pressure failures, electrical failures, and waste treatment systems;
j) fire suppression devices and alarms;

k) communication systems.

5.4 Cleaning and disinfecting procedures should be validated and documented. Only procedures that have been proven effective in inactivating poliovirus should be employed. Manufacturers are urged to develop and implement monitoring procedures for determining the disinfection of poliovirus contamination of work surfaces.

6. PRODUCTION

Production of IPV involves handling large volumes of concentrated preparations of live wild polioviruses. The majority of operations are carried in closed systems. Nevertheless leaks can occur from valves or during procedures such as taking samples for testing purposes. Effective containment therefore requires that all aspects of production, from the specifications for the facility and equipment through to personnel and working procedures, must be in compliance with each of the relevant sections of this guideline.

Personal protection and equipment

6.1 Personnel entering the production area must meet all the established requirements for entry and shall be restricted to those persons required to meet program and support needs. They shall be fully knowledgeable in all operating practices, emergency procedures, biohazards and other hazards associated with the work.

6.2 Personnel must be provided with the facilities and equipment required to maintain adequate standards of good microbiological practice and personal hygiene. Provision must be made for the changing of clothing and emergency decontamination of personnel in the event of a major spill or other release of infectious materials.

6.3 Impervious gloves must be worn at all times in the containment area and discarded as waste for decontamination when leaving the facility. Double gloving is recommended. Outer gloves must be removed and discarded after handling potentially infectious materials. Where double gloves are not worn, staff must discard gloves after handling potentially infectious materials, disinfect hands using an adequate procedure, and put on new gloves.

6.4 Hands must be washed and disinfected upon leaving the containment area. Hand-washing sinks equipped with automatic (hands free) controls and a disinfectant shown to be effective against poliovirus should be installed in the personnel air lock. All sinks must be connected with a validated waste decontamination system. The alternative use of water-free (chemical) hand-washing systems with liquid waste decontamination is acceptable.
6.5 A full body shower should be available within the personnel exit airlock from the containment area. Showers should be taken after spills, in accord with the facility plans guidelines for responding to biosafety emergencies and incidents. The biosafety committee may decide, after completion of a risk assessment that certain personnel are required to shower upon each exit from the containment area. Shower drains must be connected with a validated waste decontamination system.

6.6 An eyewash station should be available within the personnel exit airlock and on locations according to a risk assessment by the biosafety committee. Wastewater from eyewash stations within the production facility must be connected to the liquid effluent treatment system.

6.7 All laboratory clothing must be sterilized by a validated procedure before reuse or disposal.

6.8 Good microbiological practices should be rigorously enforced. These include but are not limited to:

   a) no eating, drinking, smoking, and applying of cosmetics in the containment area;

   b) no mouth pipetting;

   c) implementing policies for the safe handling of sharps;

   d) decontaminating work surfaces at least once a day and after any spill of viable material;

   e) decontaminating equipment before removal from the facility for repair or maintenance.

6.9 Data sheets and associated materials that have been used in live virus areas must be disinfected upon exit from the containment facility, or an electronic data gathering and transmission systems implemented to transfer data from the containment area.

*Polio strains for BSL-3/polio containment*

6.10 All wild type strains of live poliovirus are to be contained within BSL-3/polio biosafety conditions, including strains commonly utilized for IPV production (e.g. Mahoney, MEF-1, Saukett, Brunhilde, and Brunender strains)
6.11 Attenuated poliovirus strains (such as Sabin strains) that have been approved by the national regulatory agency in the country of manufacture for use as an oral polio vaccine, when used for manufacturing IPV, do not require containment in BSL-3/polio facilities provided they are produced under conditions that would make them suitable for oral vaccine use. If conditions other than those approved for production of oral polio vaccine are used (e.g. different multiplicity of infection or fermentation temperatures), it must be verified that the virus so produced poses no greater medical risk than that accepted for oral poliomyelitis vaccine, or BSL-3/polio containment must be instituted.

6.12 Materials containing or potentially contaminated with live poliovirus may be removed from BSL-3/polio containment conditions when the following conditions are met:

a) the material is decontaminated using a validated process proven to be effective in inactivating poliovirus, such as sterilization in an autoclave, or fumigation; or

b) in the case of in-process or quality control samples, materials have been sealed in leak proof, unbreakable, wrapped containers appropriate for the containment of pathogenic organisms for transport between production and quality control areas, and the containers have been sealed in protective wrapping (e.g. a double bag) and the outside of the container disinfected within the poliovirus containment areas; or

c) a test for inactivation of poliovirus as described for IPV production and approved by the national regulatory authorities has been completed and the results demonstrate that no residual live virus is present; or

d) following a validated inactivation procedure a kinetic measurement of virus inactivation has been completed with at least three time points, with the results of the last two time points indicating that there is no detectable live virus, and a measurement of the formalin concentration in each inactivation container has been conducted and the results indicate that the minimum validated concentration of formalin for inactivation is present. If materials are to be transferred thereafter to an area outside the BSL-3/polio containment area until such time as all tests of effective inactivation are completed, they must be stored in sealed, leak proof, unbreakable secondary storage containers.

6.13 Blending, mixing, and formulation of IPV should not be conducted using virus preparations prior to the conclusion of all tests designed to verify inactivation.
7. QUALITY CONTROL

The risks from live poliovirus in testing facilities will be different to those in the production facility. While the volumes of virus are smaller than in the production facilities, there are many more manual manipulations of samples and infected cell cultures containing viable polioviruses that are not contained in closed systems. In cases of multipurpose quality control laboratories, personnel and materials may move more frequently in and out of the BSL-3/polio containment area than is the case in a dedicated production facility. The risk assessment should reflect these important differences.

7.1 Quality control testing laboratories should maintain BSL-3/polio conditions for all areas where materials potentially infected with live poliovirus are manipulated.

7.2 Prior to using the laboratory to test other products not under BSL-3/polio containment, or when personnel not qualified to handle poliovirus are to be admitted, the laboratories must be decontaminated by a validated fumigation procedure.

7.3 In cases where quality control laboratories are housed within the production facility to enhance containment control, they must be kept separate from the production rooms, with separate air handling systems and dedicated personnel and material airlocks provided from access corridors.

7.4 Poliovirus quality control laboratories should be equipped with facilities for hand washing and disinfection. Persons must disinfect their hands after handling infectious materials, after removing gloves, and when they leave the laboratory. If sinks are used, the waste water should be collected in a waste disposal tank and disinfected prior to disposal. The alternative use of water-free (chemical) hand-washing systems with liquid waste decontamination is acceptable.

7.5 Control cell cultures for testing for adventitious agents should be considered to be potentially contaminated with poliovirus and tests conducted under biosafety 3/polio containment unless:

a) the cells have been grown in an area where there has been no poliovirus and no physical connection exists between this area and an area containing live poliovirus; or

b) samples have been taken from closed vessels that have been sterilized immediately preceding the introduction of poliovirus-free cells and growth media, and the cells and media must have been introduced into the vessel through steam-through valves or a sterile tubing weld system. In this case, the cell control laboratory must have a separate supply and exhaust air handling system, which does not circulate air to other areas, and in the
case that poliovirus cytopathic effects are observed on the control cells, the laboratory must be able to be immediately sealed and fumigated.

7.6 Samples within the containment areas should be unwrapped in the quality control testing laboratories within an appropriate biological safety cabinet only after visual inspection indicates that no leaks have occurred during transport. Wrapping should be disposed of as biohazardous waste.

7.7 Tests conducted using manual manipulations of live poliovirus on growth permissive cell substrates should be considered as high risk activities, and maintained within the biological safety cabinet to the greatest extent possible. Transfers of cell cultures infected with poliovirus between areas of the laboratory should be conducted with special care, and spills or accidents require an immediate and adequate response.

7.8 Special consideration should be given to providing adequate space within the containment area for storage of samples that may contain live poliovirus.

8. EMERGENCY PROCEDURES

Production of IPV using wild polioviruses under BSL-3/polio containment requires planning for emergencies that could result in release of live virus within the facility or into the surrounding environment. Failures of containment systems within the facility as well as external events not under the control of the manufacturer could result in the exposure of plant personnel or the public to infectious poliovirus. Response and contingency plans must be established to minimize the impact and consequences of such accidents, and adequate mechanisms must be put in place to ensure that there is a prompt and effective implementation of these plans should an incident occur.

8.1 The response to an uncontrolled release of wild poliovirus resulting from a failure in containment systems should be planned and rapidly implemented to limit exposure of persons to the virus and ensure that no further threat of exposure exists. Detailed provisions should also be made to respond to unlikely occurrences such as medical emergencies, fire, earthquake, explosions, inclement weather and extended power failures, or following access to the facility by unauthorized intruders. Special attention should be paid to events that may require the assistance of emergency personnel who may not be familiar with the facility or the infectious nature of the agents under production.

8.2 In cases of a large-scale release of poliovirus, staff and emergency personal should be supplied with protective equipment (e.g. respirators, coveralls, gloves etc.) prior to entering BSL-3/polio production and quality control units. This equipment should be available in sufficient quantities at the entrance to
the facilities, kept in good working order, and personnel should be instructed in their use.

8.3 Detailed written procedures should be available at the workplace on procedures to follow after an accident or spill involving the potential contamination of the workplace or personnel with live poliovirus.

8.4 All spills and accidents must be promptly reported to the facility manager or the biosafety officer.

8.5 Emergency equipment such as disinfectants and other clean-up materials for spills should be maintained for use in sufficient quantities to respond to the release of infected material equivalent to the maximum capacity of the facility.

8.6 The area surrounding small spills may be inactivated using adequate concentrations of validated disinfecting agents proven to be effective in killing poliovirus. Such spills would include limited leakage from valves, pipettes, small containers, or accidental dropping of culture flasks and plates in quality control laboratories. Inactivation procedures should be undertaken immediately following detection of the spill. Special attention must be paid to any procedures that may have generated aerosols.

8.7 Larger spills, such as breeches in fermentation or liquid waste vessels or explosions, should trigger an immediate assessment of the magnitude of potential contamination. The perimeter for contamination control activities should be large enough to minimize any further spread of poliovirus. The immediate response to a spill should be to evacuate the premises and return with clean-up personnel no sooner than 30 minutes after the incident, to allow aerosols to settle.

8.8 Special attention should be paid to any potential contamination of floors, walls, ceilings, equipment, airlocks, or plant or outside clothing that may have occurred as a result of the spill. Provisions should be made to verify that all areas have been decontaminated during the clean up procedure.

8.9 Personnel in the containment area at the time of the spill, emergency response personnel law enforcement, medical or fire fighting personnel, and persons involved in the risk assessment, clean-up and disinfecting of the area should all be considered at risk for a further breach in containment and subsequent poliovirus dissemination to the environment. Emergency personnel should be immunized against poliomyelitis and have adequate training to understand the need for the containment measures in place. Whenever these precautions are not possible, emergency personnel must be supplied with adequate protective clothing and equipment to ensure that they do not become infected with poliovirus in the course of their duties. Such protective clothing and
equipment must be adequately disinfected before exiting the BSL-3/polio facility.

8.10 Appropriate medical evaluation surveillance and treatment should be provided following spills. Particular caution should be exercised in monitoring potentially infected personnel for faecal shedding of poliovirus.

8.11 A full evaluation should be carried out after any emergency event involving a breach of containment. The incident and all aspects of the response to the incident should be fully investigated and documented, and revisions made to existing contingency plans and staff training as necessary to minimize its repetition.

8.12 Any major spill, accident, or suspected or confirmed poliovirus infection occurring in the area surrounding an IPV testing or manufacturing facility should be considered an urgent international public health emergency. National public health officials and responsible officials at the World Health Organization should be notified without delay.

GLOSSARY

Aerosol: A dispersion of solid or liquid particles of microscopic size in a gaseous medium.

Air balance: The necessity to keep air supply and exhaust systems in balance by means of measurements of static pressure, fan and motor performance, and air volumes.

Airlock: Areas found at entrances or exits of rooms that prevent air in one space from entering another space. These generally have two doors and a separate exhaust ventilation system. In some cases a multiple-chamber airlock consisting of two or more airlocks joined together is used for additional control.

Biocontainment technologies: The science of measurement or testing of the capability of primary and secondary containment devices.

Biosafety Committee: An institutional committee of individuals versed in the subject of containment and handling of infectious materials.

Biosafety Level (BSL)-3/Polio: A biosafety level for the containment of poliovirus with specialized air handling systems, waste effluent treatment, immunization of staff, specialized training, and validation and documentation of physical and operational requirements.

Biosafety Manual: A comprehensive document describing the physical and operational practices of the laboratory facility with particular reference to infectious materials.
Biosafety Officer: A staff member of an institution who has expertise in microbiology and infectious materials, and has the responsibility for ensuring the physical and operational practices of various biosafety levels are carried out in accordance with the standard procedures of the institution.

Backflow prevention device: A device designed to prevent backflow or back siphoning in a piping system.

Biohazard sign: A sign posted to provide information on infectious agents in use in the laboratory. Information includes the universal biohazard symbol, the name of the agent, immunization requirements for entry to the laboratory, and emergency response information.

Biological indicators: The use of organisms to test the efficacy of sterilization processes.

Biological safety cabinet: Primary and partial containment work enclosure used for manipulation of materials that may cause infections or sensitisation to workers. They are equipped with high efficiency particulate air (HEPA) filters and may or may not be open fronted.

BSI: British Standards Institute.

Certification: Documentation that a system qualification, calibration, validation, or revalidation has been performed appropriately and the results are acceptable.

Decontamination: A process by which and object or material is freed of contaminating agents.

DIN: Deutsches Institut fuer Normung e.V.

Double gloving: The wearing of two pairs of protective gloves, one over the other.

Electronic data gathering and transmission systems: Systems of recording and transmitting information such as facsimile (fax) or computer scanning systems.

Eyewash station: A dedicated device supplying tempered potable water for emergency cleansing of eyes contaminated with biological or chemical agents.

Filter housings: Airtight containment enclosures for the location of HEPA or high efficiency filters.

Floor dams: Purpose-built elevations to enclose liquid spills.

Fumigation: The process whereby gaseous chemical is applied to an enclosed space for the purpose of sterilizing the area.
Good Manufacturing Practices: That part of quality assurance which ensures that products are consistently produced as controlled to the quality standards appropriate to their intended use and as required by the marketing authorization.

HEPA filter: A filter capable of removing at least 99.97% of all particles with a mean aerodynamic diameter of 0.3 micrometers.

Inactivation: To render an organism inert by application of heat, or other means.

Magnetic badge controller: A programmable device, which will interact with door locks to allow only authorized entry to restricted areas.

Master seed lot: A culture of micro-organism distributed from a single bulk container in a single operation, in such a manner as to ensure uniformity and stability and to prevent contamination.

Mucosal immunity: Host’s immune defence mechanisms associated with mucosal surfaces.

Neutralizing antibody: Antibody, which alone or in concert with complement, neutralizes the infectivity of a virus. The level of serum neutralizing antibody which protects against clinical illness has not been determined. Titers of 1:8 are generally assumed to provide protection against contracting poliomyelitis, although it is possible that persons with low but detectable serum antibody may become re-infected with poliovirus.

NSF: National Sanitation Foundation

Penetrations: Openings through wall, floors, or ceilings to allow for mechanical services.

Positive pressure laminar flow hood: An enclosure with unidirectional outflowing air, generally used for product protection.

Primary containment: A system of containment, usually a biological safety cabinet or closed container, which prevents the escape of a biological agent into the immediate working environment.

Respirator: A respiratory protective device with an integral perimeter seal, valves and specialized filtration, used to protect the wearer from toxic fumes or particulates.

Risk analysis: A formalized documented process for analysing risks.

Secondary containment: A system of containment, usually involving specialized air handling, airlocks, and secure operating procedures, which prevents the escape of a biological agent into the external environment or into other working areas.
**Sharps:** Devices used in the laboratory, which are capable of cutting or puncturing skin (e.g., needles, scissors, glass etc.)

**Sterilization:** Sterility is the absence of viable micro-organisms. In general, an item is assumed to be sterile if the validation of the sterilization process applied to it indicates that only one item in one million items subjected to the process will contain a viable micro-organism.

**Validation:** The documented act of proving that any procedure, process, equipment, material activity, or system actually leads to the expected results.

**BIBLIOGRAPHY**


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The current version was prepared by the Secretariat and Mrs Mary Ellen Kennedy (as above) taking into account the comments from the consultation.

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