Regulations on Handling Microorganisms in Pathogenicity Groups 3, 4

[Text of Russian Federation Chief Health Inspector Order No. 4, Signed by G.G. Onishchenko, Dated 28 January 2008, and Titled "On Approval of Sanitary and Epidemiological Regulations [SP] 1.3.2322-08" with revisions dated 2 June 2009]

Russian Federation Chief Health Inspector Order No. 4, Dated 28 January 2008 and Titled "On Approval of Sanitary and Epidemiological Regulations [SP] 1.3.2322-08" (with revisions dated 2 June 2009)

In accordance with Federal Law No. 52-FZ, which is dated 30 March 1999 and titled "On the Sanitary and Epidemiology Welfare of the Population" (*Collected Legislation of the Russian Federation,* 1999, No. 14, Article 1650; 2002, No. 1 (Part 1), Article 1; 2003, No. 2, Article 167; No. 27 (Part 1), Article 2700; 2004, No. 35, Article 3607; 2005, No. 19, Article 1752; 2006, No. 1, Article 10; No. 52 (Part 1), Article 5498; 2007, No. 1 (Part I), Article 21, 29; No. 27, Article 3213; No. 46, Article 5554; No. 49, Article 6070) and Russian Federation Government Decree No. 554, which is dated 24 July 2000 and titled "On Approval of the Policy on the Russian Federation State Public Health and Epidemiology Service and the Policy on State Public Health and Epidemiological Standardization" (*Collected Legislation of the Russian Federation,* 2000, No. 31, Article 3295; 2005, No. 39, Article 3953), I order [the following]:

1. The sanitary and epidemiological regulations "Safety in Working With Microorganisms in Pathogenicity (Hazard) Groups 3 and 4 and Pathogens of Parasitic Diseases. SP 1.3.2322-08" (Attachment) shall be approved.

2. SP 1.3.2322-08 shall take effect on 1 May 2008.

G.G. Onishchenko

Registered in the Russian Federation Ministry of Justice on 21 February 2008

Registration No. 11197

Attachment: Sanitary and Epidemiological Regulations [SP] 1.3.2322-08 "Safety in Working With Microorganisms in Pathogenicity (Hazard) Groups 3 and 4 and Pathogens of Parasitic Diseases" (with revisions dated 2 June 2009)

1. Scope

1.1. These sanitary and epidemiological regulations (henceforth sanitary regulations) have been developed in compliance with Federal Law No. 52-Z, which is dated 30 March 1999 and titled "On the Sanitary and Epidemiological Welfare of the Population" (*Collected Legislation of the Russian Federation,* 1999, No. 14, Article 1650; 2002, No. 1 (Part 1), Article 1; 2003, No. 2, Article 167; No. 27 (Part 1), Article 2700; 2004, No. 35, Article 3607; 2005, No. 19, Article 1752; 2006, No. 1, Article 10; No. 52 (Part 1), Article 5498; 2007, No. 1 (Part I), Article 21, 29; No. 27, Article 3213; No. 46, Article 5554; No. 49, Article 6070) and Russian Federation Government Decree No. 554, which is dated 24 July 2000 and titled "On Approval of the Policy on the Russian Federation State Public Health and Epidemiology Service and the Policy on State Public Health and Epidemiological Standardization" (*Collected Legislation of the Russian Federation,* 2000, No. 31, Article 3295; 2005, No. 39, Article 3953).

1.2. The sanitary regulations establish the requirements regarding organizational sanitary and epidemic control (preventive) measures oriented toward ensuring personal and public safety and protection of the environment during work with group 3 and 4 pathogenic biological agents (henceforth group 3 and 4 pathogenic biological agents or pathogenic biological agents) -- microorganisms and helminths that are pathogenic for humans -- as well as all objects and materials, including field, clinical, and autopsy materials that are suspected of containing the pathogenic biological agents in question.

1.3. These sanitary regulations are intended for legal entities regardless of their organizational and lega l structure or form of ownership and also for individual businesspersons in the territory of the Russian Federation who are working with objects and materials containing or suspected of containing group 3 and 4 pathogenic biological agents.

As per Russian Federation Chief Health Inspector Decree No. 42, dated 2 June 2009, changes taking effect on 1 August 2009 shall be inserted in Paragraph 1.4 of this attachment.

See the text of the paragraph in the previous edition.

1.4. Compliance with the requirements in these sanitary regulations shall be mandatory for legal entities for individual businesspersons who are working with pathogenic biological agents:

Group 3:

- Diagnostic (for the purpose of pathogen detection and isolation and for experimental and production work);
- Polymerase chain reaction [PCR] diagnosis;
- Diagnostic tests for cholera and botulinum toxin that are being performed for purposes of preventing these infections;
- Immunologic tests with group 3 pathogenic biological agents;
- Immunologic tests to detect (in people's blood) antigens to pathogenicity group 2 microorganisms (without accumulation of the pathogen) and/or antibodies against them;
- Experimental and production work with vaccine strains of pathogens that belong to pathogenicity group 1 or 2 or are officially classified as belonging to group 3; and
- Tests to monitor environmental objects and product quality.

Group 4:

- Diagnostic (for the purpose of pathogen detection and isolation and for experimental and production work);
- Immunologic tests involving pathogenic biological agents belonging to group 3 (without accumulation of the pathogen);
- Tests for the presence of sanitary indicator microorganisms to monitor environmental objects and product quality and products' quality; and
- PCR testing.

II. Requirements regarding organizing work with group 3 and 4 pathogenic biological agents

As per Russian Federation Chief Health Inspector Decree No. 42, dated 2 June 2009, changes taking effect on 1 August 2009 shall be inserted in Paragraph 2.1 of this attachment.

See the text of the paragraph in the previous edition.

2.1. General requirements

2.1.1. The activity of legal entities regardless of their organizational and legal structure or form of ownership and individual businesspersons related to use of group 3 and 4 pathogenic biological agents and pathogens of parasitic diseases must be conducted in compliance with Federal Law No. 128-FZ, which is dated 8 August 2001 and titled "On Licensing Individual Types of Activity" (*Collected Legislation of the Russian Federation,* 2001, No. 33 (Part 1), Article 3430; 2002, No. 11, Article 1020; 2002, No. 50, Article 4925; 2003, No. 2, Article 169; 2003, No. 9, Article 805; 2003, No. 11, Article 956; 2003, No. 13, Article 1178; 2003, No. 52 (Part 1), Article 5037; 2004, No. 45, Article 4377; 2005, No. 13, Article 1078; 2005, No. 27, Article 2719; 2006, No 1, Article 11; 2006, No. 31 (Part 1), Article 3455; 2006, No. 50, Article 5279; 2007, No. 1 (Part 1), Article 7; 2007, No. 1 (Part 1), Article 15; 2007, No. 7, Article 834; 2007, No. 30, Article 3748; 2007, No. 30, Article 3749; 2007, No. 30, Article 3750; 2007, No. 45, Article 5427; 2007, No. 46, Article 5554; 2007, No. 49, Article 6079; 2007, No. 50, Article 6247; 2008, No. 18, Article 1944; 2008, No. 29 (Part 1), Article 3413; 2008, No. 30 (Part 1), Article 3604: 2008, No. 30 (Part 2), Article 3616; 2008, No. 52 (Part 1), Article 6227; 2009, No. 1, Article 15).

2.1.2. The activity of each structural subunit (microbiology laboratory, shop, production section, and so forth) connected with use of group 3 and 4 pathogenic biological agents must be conducted on the basis of a sanitary and epidemiological certificate in accordance with the Federal Law "On the Sanitary and Epidemiological Welfare of the Population."

The second paragraph has been eliminated.

See the text of the section paragraph of 2.1.2.

2.1.3. The tracking, storage, transfer, and transport of group 3 and 4 pathogenic biological agents must be conducted in compliance with the existing sanitary and epidemiological regulations.

Transfer of group 3 and 4 pathogenic biological agents to organizations that are not licensed for activity connected with use of infectious disease-causing pathogens belonging to the respective pathogenicity groups shall not be permitted.

Group 3 and 4 pathogenic biological agents shall be stored in a "contaminated" zone. In some cases, with the consent of bodies conducting state public health and epidemiological surveillance, their storage in a specially isolated and equipped "clean zone" shall be permitted provided they are packed in accordance with the requirements established for transporting group 3 and 4 pathogenic biological agents.

2.1.4. Work with recombinant molecules of the DNA of group 3 and 4 pathogenic biological agents shall be regulated by Federal Law No. 86-FZ, which is dated 5 July 1996 and titled "On State Regulation in the Area of Genetic Engineering Activity" (*Collected Legislation of the Russian Federation,* 1996, No. 28, Article 3348; *Rossiyskaya Gazeta*, No. 135, 14 July

2000) and by normative documents on safety when working with recombinant DNA molecules and existing sanitary and epidemiological regulations.

2.1.5. Work related to production of medical immunobiological preparations that involves use of group 3 and 4 pathogenic biological agents shall be regulated by these sanitary regulations and by other normative documents containing requirements regarding premises, equipment, safety engineering, and industrial sanitation.

2.1.6. [The following tests] may be performed in laboratories that work with microorganisms in pathogenicity group 3: tests for cholera and botulinum toxin that are being conducted for the purpose of preventing cholera and botulism; immunologic (serological) tests to detect antigens of microorganisms of pathogenicity group 2 in people's blood (without accumulation of the pathogen) and/or antibodies against them; and PCR tests (without accumulation of the pathogen) to detect pathogens of brucellosis, parenteral hepatitis B and C viruses, AIDS, and other microorganisms in pathogenicity group 2 that are regulated by existing normative and methodological documents. Immunologic (serological) tests and PCR tests shall be conducted in a biohazard room or in a biohazard hood.

2.1.7. Each structural subunit conducting work with group 3 and 4 pathogenic biological agents shall have a document developed for it that specifies the procedure for safe operation under the specific conditions with consideration for the nature of the work, features of the process, properties of the microorganism, and the products of its vital functions. The safety requirements must be no lower than the requirements stipulated by these sanitary regulations. The document must be reconciled with the committee for monitoring compliance with the organization's biological safety requirements and approved by its head.

When developing and/or introducing new methods and methodological techniques requiring strengthening of safety measures, the appropriate additions shall be added to the document.

2.2. Requirements regarding establishing personnel's clearance for work with group 3 and 4 pathogenic biological agents and regarding medical observation of personnel

2.2.1. Work with group 3 and 4 pathogenic biological agents may be performed by specialists who are at least 18 years of age and have the higher or secondary medical, biological, veterinary, or other education appropriate to the procedure for filling jobs that has been approved by each department and who have completed the appropriate courses of specialization (including mastery of methods for safe handling of gro up 3 and 4 pathogenic biological agents) and do not have any contraindications to vaccination, treatment with specific drugs, or working while in personal protective gear.

2.2.2. Clearance of personnel for work with group 3 and 4 pathogenic biological agents shall be granted on the basis of an order of the head of the organization that is issued once every 2 years with consideration for point 2.2.1 of this section and on the basis of verification of the personnel's knowledge of biological safety requirements. Training on compliance with biological safety requirements must be conducted at least once each year.

As per Russian Federation Chief Health Inspector Decree No. 42, dated 2 June 2009, point 2.2.3 of this attachment shall be stated in a revision that will take effect on 1 August 2009.

See the text of this point in the previous edition.

2.2.3. Engineering and technical personnel, disinfectors, and medical assistants of a subunit

whose activity entails use of group 3 and 4 pathogenic biological agents must complete introductory and periodic on-the-job training in biological safety in accordance with his job duties. Engineering and technical personnel shall receive clearance to service equipment based on an order of the organization's head once every 2 years.

2.2.4. Permission for engineering and technical personnel who are not regular employees of the organization to visit laboratories, shops, sections, or a specific workstation in order to service equipment shall be issued by the subunit's manager. The visit must be made under the escort of an employee of the structural unit after work as been halted and disinfection has been conducted. The visit must be recorded in a special log.

2.2.5. Specialists who are not permanently employed at an organization may be allowed to work with group 3 and 4 pathogenic biological agents on the same basis as everyone else in accordance with the requirements of point 2.2.1 of this section.

2.2.6. When being hired for a job entailing use of group 3 and 4 pathogenic biological agents, personnel must undergo a preliminary medical examination for the purpose of discovering medical contraindications to vaccine prophylaxis, treatment with specific drugs, and use of personal protective gear. The scope of and procedure for conducting medical examination shall be determined by the normative documents that are in effect.

All employees employed in jobs involving group 3 and 4 pathogenic biological agents must undergo periodic medical examinations in accordance with the normative documents.

2.2.7. Employees of laboratories that perform serological tests for HIV infection and hepatitis B and C shall undergo annual monitoring tests for presence of the respective antigens (antibodies) in their blood serum.

2.2.8. Employees who work with blood (blood serum or plasma) must be immunized against hepatitis virus, and employees who perform tests for enteroviruses must be immunized against poliomyelitis.

2.2.9. In the event that an employee develops symptoms characteristic for an infectious disease caused by a pathogen with which he has worked, the employee must notify the subunit's manager of this fact.

2.3. Requirements regarding laboratory premises and equipment

2.3.1. Microbiological laboratories that work with group 3 and 4 pathogenic biological agents must be located in a separate building or in an isolated part of the building. The laboratory's entrance door must display the laboratory's name (number) and the international Biohazard sign.

Locating laboratories in residential buildings shall not be permitted.

2.3.2. Industrial laboratories that work with group 3 pathogenic biological agents must be located in separate buildings or in an isolation unit of a building that has a separate entrance, whereas industrial laboratories that work with group 4 pathogenic biological agents may be located in an isolation unit in an industrial building .

2.3.3. Diagnostic laboratories that perform tests involving group 3 and 4 pathogenic

biological agents must have two entrances: one for employees and one for delivery of material for testing. Intake of material through a pass-through shall be permitted.

Laboratories of scientific research organizations that conduct experimental research with group 3 and 4 pathogenic biological agents and industrial laboratories are permitted to have a single entrance.

2.3.4. The laboratory must be equipped with cold and hot water service, plumbing, electricity, heating, and ventilation.

All laboratory premises must have natural and artificial lighting in accordance with the requirements of the normative documents that are in effect.

2.3.5. Space-planning decisions and equipment placement must provide routes for traffic of group 3 and 4 pathogenic biological agents and personnel and also ensure compliance with the requirements in these sanitary regulations.

2.3.6. Laboratories must have a set of work and utility spaces (rooms). The selection of buildings and the equipment with which they are outfitted may vary depending on the laboratory's specific goals and tasks.

As per Russian Federation Chief Health Inspector Decree No. 42, dated 2 June 2009, point 2.3.7 of this attachment shall be stated in a revision that will take effect on 1 August 2009.

See the text of this point in the previous edition.

2.3.7. Laboratory spaces shall be divided into a "contaminated" zone (where group 3 and 4 pathogenic biological agents undergo procedures and are stored) and a "clean" zone (where microorganisms are not worked with or stored).

The "clean" zone must contain the following spaces:

- A cloakroom for outerwear;
- Spaces for preliminary operations (getting ready, washing up, preparing and dispensing culture media, and so forth);
- A space for sterilizing culture media and laboratory vessels (a sterilization area);
- A space with a walk-in refrigerator or refrigerators for storing culture media and diagnostic preparations;
- A space for working with documents and literature;
- A space for resting and eating;
- An office for the laboratory's head;
- A space for storing work clothes and putting them on;
- Utility spaces; and
- A toilet.

For work with group 3 and 4 pathogenic biological agents, the "contaminated" zone must contain the following:

- A space for receiving and registering material (samples);
- Isolation areas with a dressing area or spaces equipped with biohazard hoods.
- Spaces for performing bacteriological (virological) tests;
- A space for luminescence microscopy;
- A space for performing zoological and entomological tasks;
- A space for performing parasitologic tests;
- A space for working with laboratory animals (infection, dissection);
- A space for keeping infected laboratory animals;
- Spaces for PCR diagnosis;
- A temperature-controlled room; and
- A space for decontamination (autoclaving).

As per Russian Federation Chief Health Inspector Decree No. 42, dated 2 June 2009, point 2.3.8 of this attachment shall be stated in a revision that will take effect on 1 August 2009.

See the text of this point in the previous edition.

2.3.8. A decontamination station must be provided at the boundary between the "clean" and "contaminated" zones in laboratories that are being newly built or rebuilt.

As per Russian Federation Chief Health Inspector Decree No. 42, dated 2 June 2009, point 2.3.9 of this attachment shall be stated in a revision that will take effect on 1 August 2009.

See the text of this point in the previous edition.

2.3.9. Laboratories that perform tests solely with group 4 pathogenic biological agents must contain the following items in their "contaminated" zone:

- A room for inoculating [cultures];
- A room for performing tests on pathogenic biological agents;
- A room for decontamination and sterilization; and
- A shower in a decontamination station at the boundary between the "clean" and "contaminated" zones.

Labeling of autoclaves, tables, and shelves shall be mandatory, as shall time separation of the movement of infectious and clean materials.

2.3.10. When several microbiological laboratories are located in the same unit, they may share the following [items]: a unit for working with infected animals, autoclave units for decontamination, washrooms, rooms for preparing culture media, and other utility spaces.

As per Russian Federation Chief Health Inspector Decree No. 42, dated 2 June 2009, changes taking effect on 1 August 2009 shall be inserted in point 2.3.11 of this attachment.

See the text of this point in the previous edition.

2.3.11. The interiors of premises must be finished in accordance with their functional purpose and health regulations. The surface of the floor, walls, and ceilings in laboratory

rooms of the "contaminated" zone must be smooth, free of crevices, and resistant to repeated exposure to detergents and disinfectants. The floors must not be slippery, and they must be waterproof.

The premises of the "contaminated" zone are not permitted to have suspended ceilings that to not meet the aforesaid requirements or underfloor ducts.

2.3.12. The exposed and concealed pipes (radiators) in the premises of the "contaminated" zone shall not be located directly next to the walls so as to allow for the possibility of disinfecting them, and the places where utility lines enter must be leak-tight.

Heating devices must have a smooth and easy-to-clean surface.

2.3.13. The windows and doors of the premises of the laboratory's "contaminated" zone must be leak-tight. The window openings may be filled with glass blocks. Regardless of whether a security alarm system is present, the basement and first-floor windows must be equipped with metal grating that does not violate fire safety rules. The doors must have locking devices.

2.3.14. The entrance doors to the premises for working with infected animals must be equipped with high thresholds that cannot be infiltrated by rodents.

2.3.15. The instruments, equipment, and measuring instruments that are used in the laboratory's work must be certified and technically sound, and they must have technical documentation and operating instructions that give consideration to biological safety requirements. Measuring devices must undergo metrological inspection at the established times.

2.3.16. Scheduled preventive maintenance of laboratory equipment and engineering systems for ensuring subunits' biological security shall be conducted by the engineering and technical service and specialists in accordance with the yearly schedule.

2.3.17. Laboratory equipment and furniture (tables, shelves for keeping animals, chairs, and so forth) must be smooth as well as free of sharp edges and roughness, and they must have a covering that is resistant to the effects of detergents and disinfectants. Table surfaces must not have seams or cracks. Use of furniture that is made of wood or has a soft covering in the premises of the "contaminated" zone shall not be permitted.

2.3.18. The passages to workstations or between two rows of protruding equipment must be at least 1.5 m wide.

2.3.19. The premises of the "contaminated" zone must be equipped with bactericidal irradiation machines to decontaminate the air and surfaces in accordance with the regulations.

2.3.20. The work tables in laboratory premises must be provided with protection against direct sunlight. A light-protective film or blinds made of a material that is resistant to exposure to disinfectants may be used for these purposes.

2.3.21. Laboratory premises must be impermeable to rodents and insects.

2.3.22. Laboratory premises must be equipped with fire alarms and provided with fire extinguishers in accordance with fire safety requirements.

2.3.23. All liquid wastes that are generated during the process of work in the "contaminated" zone must undergo mandatory chemical or thermal decontamination before being released into the sewage system.

As per Russian Federation Chief Health Inspector Decree No. 42, dated 2 June 2009, point 2.3.24 of this attachment shall be stated in a revision that will take effect on 1 August 2009.

See the text of this point in the previous edition.

2.3.24. The premises of the unit for working with and keeping infected animals, isolation rooms, and microbiology rooms must be equipped with automatic positive-pressure ventilation systems with mechanical excitation that is equipped at the outlet with fine-gauge filters that are checked for their protective effectiveness or class II biohazard hoods.

In some cases, intake ventilation systems may also be equipped with fine-gauge filters to create aseptic conditions in premises.

2.3.25. The positive-pressure ventilation systems of laboratories (laboratory buildings) must be operated according to instructions that have been developed based on the requirements of the corresponding normative documents.

2.3.26. Filters must be replaced in cases of violation of the parameters of the depression mode (a change in air flow rates or the air exchange rate), when a filter is damaged (a decrease in resistance or increase in the penetration coefficient), or when the filter's resistance increases by 50% and the air flow rate in the isolation devices decreases simultaneously.

As per Russian Federation Chief Health Inspector Decree No. 42, dated 2 June 2009, point 2.3.27 of this attachment shall be stated in a revision that will take effect on 1 August 2009.

See the text of this point in the previous edition.

2.3.27. To maintain the normative microclimate parameters, air conditioners may be installed in workrooms and isolation rooms. Air conditioners must be turned off during work with pathogenic biological agents. Air conditioners' filter elements must be cleaned periodically (at least once every 3 months) to remove mechanical particles and disinfected. Placement of air conditioners in rooms used to keep infected animals shall not be permitted.

2.3.28. All vacuum lines and compressed air and gas lines in the "contaminated" zone of laboratory premises must be equipped with fine-gauge air filters.

2.3.29. Extending hot and cold water supply and sewage systems into microbiology hoods shall not be permitted.

2.3.30. To ensure physical protection of working personnel, workplace's air and surfaces, and the environment against microorganisms that are undergoing testing, biohazard hoods must be used.

As per Russian Federation Chief Health Inspector Decree No. 42, dated 2 June 2009, changes taking effect on 1 August 2009 shall be inserted in point 2.3.311 of this attachment.

See the text of this point in the previous edition.

2.3.31. Class II biohazard hoods must be used for working with pathogenic biological agents.

All work in biohazard hoods shall be performed on trays with liners that have been moistened with a disinfectant solution.

The space for tests on intestinal protozoa and helminths must be equipped with an exhaust fume hood.

2.3.2. If they cannot be performed in biohazard hoods, tasks associated with a high level of risk of aerosol formation (centrifugation, homogenization, grinding, intensive shaking, ultrasound treatment, and dissection of objects containing infected material), tasks involving large amounts and high concentrations of pathogenic biological agents, and so for th must be performed in individual isolation rooms.

2.3.33. Biohazard hoods must be checked for their protective effectiveness:

- After they have been installed and readied for use;
- At least once per year if filters for preliminary removal of coarse particles from air are present;
- At least once every 6 months if filters for preliminary removal of coarse particles from air are not present; and
- After the hood has been moved or repaired.

During the testing, the effectiveness of the air filters' operation and air flow rate in the hood's working opening must be determined.

2.4. Requirements for performing work in the laboratory

As per Russian Federation Chief Health Inspector Decree No. 42, dated 2 June 2009, changes taking effect on 1 August 2009 shall be inserted in point 2.4.1 of this attachment.

See the text of this point in the previous edition.

2.4.1. Material to be tested shall be delivered to the laboratory in containers, dressing boxes, or cool boxes. Containers holding liquid materials that are being delivered must be closed with stoppers that eliminate the possibility of the contents spilling out during transport. The bottom of containers carrying containers that are holding pathogenic biological agents must be covered with an adsorbent material (a gauze pad, fabric, absorbent cotton, or the like), and containers, boxes, or cool boxes must be labeled and have the international Biohazard sign. Delivery of material in shopping bags, suitcases, briefcases, or items intended for personal use shall not be permitted.

2.4.2. Acceptance and sorting of delivered material (samples) must be performed in compliance with precautionary measures. Containers holding pathogenic biological agents must be placed on a tray or dish covered with multiple layers of absorbent cotton that have been moistened with a disinfectant solution. Personnel must use a mask and rubber gloves.

2.4.3. The following work shall be performed in the isolation spaces of a laboratory's

"contaminated" zone (or in biohazard hoods):

- Working with animals (infection, dissection);
- Keeping infected animals;
- Centrifuging pathogenic biological agents, drying, disintegration, and performing other procedures during which aerosol formation is likely;
- Infecting a cell culture or chicken embryos;
- Preparing suspensions;
- Working with lyophilized pathogenic biological agents;
- Performing work related to maintaining collection strains; and
- Performing work related to identification and study of microorganism strains that have been isolated.

2.4.4. During work, the doors of cubicles and dressing areas must be closed. Exiting cubicles while work is underway shall not be permitted.

The cubicle must be equipped with emergency alarms systems, and the dressing area must be equipped with a fire-fighting equipment.

2.4.5. When biohazard hoods are used, ventilation should be turned on before beginning the work is begun. The direction and velocity of the air in the apertures of class II hoods are determined when they are installed and after scheduled maintenance. Before the study material is loaded into the hood's working space, a check needs to be performed to ensure that the equipment in the hood is in good working order and that a supply of disinfectant is present.

All work must be performed toward the back of the class II biohazard hood and it must be visible from the outside.

After the containers holding pathogenic biological agents have been removed, the biohazard hood's front panel shall be lowered and the bactericidal lamps inside the hood shall be turned on.

2.4.6. Infection of animals in hoods shall be performed in the presence of two people.

2.4.7. Only rubber pipette bulbs or automatic devices may be used during pipetting.

2.4.8. The bacteriological loop must be closed into a continuous ring an d have an arm no longer than 6 cm. Use of disposable commercially manufactured loops with a longer arm length shall be permitted.

2.4.9. Before a vessel, pipette, piece of equipment, syringe, or the like is used, its integrity and working order must be checked.

2.4.10. When human blood sera are tested to detect antigen or antibodies against pathogens in pathogenicity group II:

- The work shall be performed in a separate space (room, hood);

- The work shall be done using only noninfectious antigens (diagnostic preparations); and
- Separation of blood sera by centrifugation must be performed in an isolated space or biohazard hood.

2.4.11. Work related to lyophilization of group 3 and 4 pathogenic biological agents shall be conducted in accordance with the instructions that are in effect.

2.4.12. Ampules containing dried cultures shall be opened in the live cultures museum (collection) under a biohazard hood. The tapered end of the ampule shall be heated over a burner flame, after which a small piece of sterile absorbent cotton that has been moistened in sterile water shall be carefully placed in contact with it to form a crack. The same moistened absorbent cotton shall be wrapped around the ampule's spout. After a circular (or not completely circular) crack has formed, the end of the ampule shall be covered with a triple layer of gauze wadding (which has been moistened with disinfectant and squeezed out well) and snapped off with forceps. After the ampule has been opened, it shall be left covered for 1 to 2 minutes. The absorbent cotton shall then be removed and submerged (along with any pieces of glass) into disinfectant solution. The opened ampule shall be covered with a sterile gauze plug for 1 to 2 minutes, after which solution for preparing a suspension shall be added to the ampule and then used to inoculate liquid or solid culture media.

2.4.13. After the work has been completed, all objects containing pathogenic biological agents must be collected and placed in a storage devices (refrigerators, temperature-controlled chambers, cabinets, and so forth). The tables' working surfaces must then be disinfected by the mandatory procedure.

2.4.14. The used pipettes shall be completely (vertically) submerged in disinfectant, avoiding formation of air bubbles in the channels.

2.4.15. The residues of the pathogenic biological agents, used vessels, and solid wastes must be collected from the laboratory's "contaminated" zone, placed in closed containers, and either transferred to the autoclave room or disinfected on site. Pouring non-decontaminated liquids down the drain shall be prohibited.

As per Russian Federation Chief Health Inspector Decree No. 42, dated 2 June 2009, changes taking effect on 1 August 2009 shall be inserted in point 2.4.16 of this attachment.

See the text of this point in the previous edition.

2.4.16. The process of transferring the pathogenic biological agents and used vessels for disinfection must be completed in closed containers with the appropriate labeling.

As per Russian Federation Chief Health Inspector Decree No. 42, dated 2 June 2009, point 2.4.17 of this attachment shall be stated in a revision that will take effect on 1 August 2009.

See the text of this point in the previous edition.

2.4.17. Test tubes and vials containing blood clots shall be disinfected by using disinfectant solutions or by using physical disinfection methods with equipment that has been duly authorized for these purposes. Dumping a non-disinfected blood clot out of a test tube (vial) shall be prohibited. Caution must be exercised when submerging containers holding blood clots in disinfectant solution. The container shall be grasped with forceps so that one of its

jaws is slightly in the solution, and it shall be submerged at an angle until it has been completely filled with solution. When the container is submerged correctly, air bubbles do not form and the container is lowered to the bottom. After all the containers have been submerged, the forceps shall be disinfected.

2.4.18. After the work has been completed, the laboratory's "contaminated zone" shall be locked and sealed. If a collection of microorganism cultures is present, their storage unit shall also be sealed. The application and removal of seals shall be performed by employees of the laboratory who have authorization from the head of the laboratory (subunit).

2.4.19. Pathogenic biological agents shall be stored, kept track of, transferred, transported, and destroyed in accordance with the requirements in the existing regulatory documents.

2.4.20. Receiving visitors, storing food products, and receiving food shall be permitted only in specially designated locations in the laboratory's "clean" zone.

2.4.21. Laboratory equipment, laboratory vessels or tableware, reagents, instruments, and so forth shall be removed from the laboratory only after they have been disinfected and only with the permission of its head.

2.4.22. Use of personal hygiene materials and agents that irritate the skin shall not be permitted.

2.4.23. The following [actions] shall not be permitted in the laboratory's "contaminated" zone:

- Leaving unfixed smears or vessels containing pathogenic biological agents in workstations after work has ended;
- Pipetting with the mouth and pouring an infectious liquid material over the edge of a glass vessel (test tube, flask, vial, and so forth);
- Storing outerwear, headwear, footwear, umbrellas, shopping bags, cosmetics, and so forth and food products;
- Smoking and drinking water;
- Leaving a workstation while any type of work with pathogenic biological agents is being performed;
- Pouring liquid wastes (infected liquids, test material, and so forth) down the drain without first decontaminating it; and
- Removing non-decontaminated blood clots from test tubes or vials by shaking them out.

2.4.24. Successive diagnostic and experimental tests in one and the same space after the space, instruments, and equipment have been disinfected shall be permitted.

2.5. Requirements regarding production jobs

2.5.1. The procedure for working in production areas when working with cultures of microorganisms belonging to pathogenicity groups 3 and 4 shall be established in accordance with these sanitary regulations, the sanitary regulations titled "Proper Practice in Production of Medical Immunobiological Preparations. SP 3.3.2.1288-03" (registered in the Russian Federation Ministry of Justice on 22 May 2003; registration number, No. 4584), and also the instructions on freeze-drying infectious disease-causing pathogens belonging to

pathogenicity groups 1 through 4.

2.6. Additional requirements when working with hydatid and alveolar echinococci

2.6.1. Experimental work involving the strobila (tapeworm) sta ge of hydatid and alveolar echinococcosis shall be authorized for only 2 weeks from the moment of oral infection of animals with parasite protoscolices.

Work with mature eggs of the tapeworm stage of the aforesaid echinococci must be conducted in biohazard hoods (at least class II).

2.7. Additional requirements when working with tuberculosis pathogens

2.7.1. Work with tuberculosis pathogens and material suspected of being infected with tuberculosis pathogens shall be performed in accordance with Attachment 1.9-1.11 to Russian Ministry of Health Order No. 109, which is dated 21 March 2003 and titled "On Improving Tuberculosis Control Measures in the Russian Federation (no need for state registration in the Russian Federation Ministry of Justice. Russian Federation Ministry of Justice Letter No. 07/4535-YuD, which is dated 6 May 2003).

2.8. Requirements regarding performing work involving the use of aerosol chambers

2.8.1. Aerosol chambers (units) must be located in isolated spaces of the "contaminated" zone. Cubicles for keeping infected animals and dissecting them must be directly adjacent to the aerosol chamber's cubicle. All the cubicles must be connected by means of pass-throughs.

2.8.2. Cubicles for housing an aerosol chamber, keeping animals, and dissecting them must be equipped with mechanical negative-pressure ventilation with fine-gauge air filters and must have a redundant motor on the exhaust fan that switches on automatically.

It must support the discharge of a 2- to 4-mm water column, or the exhaust must be at least 15% stronger than the intake.

2.8.3. After installation, air filters must be checked for penetration, and their resistance must be measured.

2.8.4. During operation, filters' resistance must be measured quarterly, and the readings must be recorded in a special log.

2.8.5. Filters must be replaced in cases of violation of the parameters of the depression mode (a change in air flow rates or the air exchange rate), when a filter is damaged (a decrease in resistance or increase in the penetration coefficient), or when the filter's resistance increases by 50% and the air flow rate in the isolation devices decreases simultaneously.

2.8.6. The aerosol chamber's design must ensure constant discharge of at least a 4-mm water column within it, and it must be equipped with an air cleaning (decontamination)

system.

2.8.7. Testing (by using test microbes) of the aerodynamic unit for the possibility of penetration of the aerosol into the air of the space must be checked annually.

2.8.8. Ventilation activation buttons must be equipped with an indicator light.

2.8.9. The interior finish of the cubicles for housing the aerosol chamber and for keeping and dissecting animals (floors, walls, and the ceiling) must withstand systematic aerosol disinfection.

2.8.10. Work in an aerosol chamber that involves infected animals must be performed in a type 4 hazmat suit (loose-fitting trousers or coveralls, coat, socks, slippers, and cap), as well as with gloves and cotton gauze masks or respirators of the Lepestok-200 [*Translator's note*: a type of filtering respirator] type.

2.8.11. Protective clothing shall be removed and soaked in disinfectant solution in the dressing area.

2.8.12. Before each session of work involving the aerosol unit, the unit and ventilation system must be inspected and their readiness for the work must be determined.

2.8.13. In each subunit, detailed instructions on the procedure for working in an aerosol unit and with infected animals that give consideration to biological safety requirements and that also specify the measures to be implemented when localizing and eliminating acciden ts must be compiled and approved by the institution's head.

2.9. Requirements regarding the procedure for trapping, transporting, and keeping wild vertebrate animals and arthropods

2.9.1. The procedure for trapping, transporting, removing, and keeping wild vertebrate animals and arthropods in areas where plague, hemorrhagic fevers, and other extremely dangerous natural focal infections are enzootic shall be specified in the sanitary regulations regarding working safely with microorganisms in pathogenicity groups 1 and 2.

2.9.2. In an area where plague and other extremely dangerous natural focal infections are not enzootic, vertebrate animals and blood-sucking arthropods shall be kept in strict compliance with these regulations.

2.9.3. Before work related to trapping wild vertebrate animals and arthropods is begun, the head of the epidemiological team (expedition) must obtain a certificate (from the regional body of the Federal Service for Surveillance in the Area of Protection of Consumer Rights and Human Welfare [Rospotrebnadzor]) confirming that the area where trapping is planned has not had any epizootics or human cases of natural focal infections during the previous 3 years.

2.9.4. The leader (head) of the epidemiological team (expedition) shall be responsible for compliance with biological safety regulations during work with pathogens of natural focal infections that are circulating in the specified area.

2.9.5. Before they are transported to research and other organizations, wild animals and arthropods that are trapped in the wild shall be held under quarantine. A quarantine vivarium may be set up at the base of the temporary epidemiological team (expedition) or at a

standing organization. The quarantine shall last for 1 month.

2.9.6. The premises of the quarantine vivarium and the insectarium must be isolated from other spaces and protected against penetration by rodents and insects.

2.9.7. The head of the epidemiological team (expedition) shall be responsible for compliance with biological safety regulations in the quarantine vivarium and in the space for working with arthropods.

2.9.8. Wild animals that are brought to the quarantine vivarium must be free from arthropods and must be placed in clean metal or glass containers with solid screen covers.

2.9.9. In the event of discovery of a dead animal, bacteriologic (virologic) and serological tests must be performed on its carcass.

2.9.10. If an infectious or parasitic disease is discovered among the animals, their quarantine period must be extended for 1 month from the date on which the last animal death was recorded. In the event of mass death, all the animals shall be killed and the vivarium shall be carefully disinfected.

2.9.11. The carcasses of the dead or killed animals shall be decontaminated.

2.9.12. After the quarantine period has ended, the healthy animals shall be transported to the location where they are to be used.

2.9.13. Arthropods shall be kept in a special space (insectarium) in cages or containers that rule of the possibility of their scattering.

2.9.14. Vessels and instruments used in working with arthropods shall be disinfected.

2.9.15. The movements of wild vertebrate animals and arthropods in the vivarium and insectarium shall be recorded in a special log, and the location and date of their trapping and results of their testing and quarantine shall be noted.

2.9.16. Transfer of wild vertebrate animals and arthropods from the vivarium and insectarium to other institutions is possible only with permission of the organization's head. Only those animals that were born in a clean vivarium may be dispensed.

2.10. Requirements regarding performing zoologic or entomologic work

2.10.1. Implements used in trapping and other tools that come into contact with rodents and arthropods during the process of working with them (traps, mashers, strips for collecting ectoparasites, test tubes, bags, and so forth) must be transported or hand carried in a closed container. Implements for catching and collecting field material shall be brought to the laboratory in a specially designated vehicle under the escort of an individual who is familiar with biological safety requirements.

Implements for catching and collecting field material need to be stored in specially designated locations that cannot be accessed by outsiders.

2.10.2. When necessary, wild animals that have been obtained shall be killed directly in the trap by compressing their neck with forceps or crucible tongs. For safety during

transportation, the carcasses shall be placed in coarse calico bags, and the latter shall be placed in transport containers, chests, or canvas (oilskin) bags. To prevent arthropods from scattering, the coarse calico bags shall be tied tightly twice (the second time through the folded-under edge of the bag) and delivered to the laboratory for testing.

2.10.3. Live rodents sh all be placed in metal transport containers or in transport containers or chests that have been lined on the inside with galvanized iron. Arthropods for parasitologic and microbiological tests shall be delivered in test tubes covered with cotton gauze stoppers that have been placed in metal cases or in thick-walled glass vials with plug stoppers that have been placed in coarse calico bags.

2.10.4. After they have removed from their bags, rodents that have been delivered dead shall be stripped, and rodents that have been delivered alive shall be treated with insecticides while in their transport containers.

2.10.5. The coarse calico bags used to deliver wild animals and other material shall be subjected to insect extermination and disinfection after each use (application).

2.10.6. Trapping implements and other tools shall be disinfected daily at the end of the workday.

2.10.7. Determination of arthropods' species and laboratory testing (preparing a suspension and inoculating it) shall be performed in a space for zoological and entomological work. Before the determination, arthropods shall be immobilized with ether vapors, placed on a wide glass slide, and examined under a microscope while dry.

When live arthropods are examined in a drop of water under a cover glass, the glass slide shall be placed in a Petri dish to rule out the possibility of the microscope stage being contaminated by liquid draining off of the glass. After the work has been completed, the Petri dishes and glass shall be submerged in disinfectant solution. To avoid splashing of the liquid when suspensions of ticks are being prepared, they need to be cut from the Petri dish or large funnel by using scissors under protection of a covering.

2.10.8. The procedure for removing pelts from wild animals that were captured in areas where an epizootic was possible or in progress and preparing carcasses for collections shall be as follows:

2.10.8.1. When preparing collection carcasses for instructional purposes, the wild animals need to first be kept in a 10% formalin solution. The exposure time shall be determined by proceeding from the size of the wild animal and the rate at which formalin penetrates the tissue (1 cm/day). Work with wild animals that have been fixed in formalin may be performed in any working space. The protective clothing shall not be regulated.

2.10.8.2. When preparing carcasses for research purposes (when exposure to formalin is not permissible), the wild animal shall be submerged for 10 to 15 minutes in a 5% solution of Lysol before removal of its pelt, and the removed pelt shall be submerged in a Lysol solution again for 3 hours, after which the fat shall be removed from it, it shall be washed, and its inner surface shall be treated with sodium arsenide. The skull shall either be kept in sodium arsenide or disinfected by boiling. Removal of pelts from rodents shall be performed in a space for working with infected animals in compliance with biological safety requirements.

2.11. Requirements regarding the procedure for using work clothes and personal protective

gear

2.11.1. Laboratory personnel must be provided with work clothes: lab coats, loose-fitting trousers (coveralls), caps, disposable footwear, and personnel protective gear depending on the nature of the work being performed and in accordance with existing norms.

2.11.2. Work clothes and footwear must be individually tailored and conform to workers' size, and they must be stored separately from personal clothing.

2.11.3. When tests are performed in isolated locations, the lab coat must be replaced by a hazmat suit or surgical scrubs that extend to the lower third of the shin. In addition, rubber gloves, slippers, and (when necessary) respirators (masks) shall be used.

As per Russian Federation Chief Health Inspector Decree No. 42, dated 2 June 2009, changes taking effect on 1 August 2009 shall be inserted in point 2.11.4 of this attachment.

See the text of this point in the previous edition.

2.11.4. In addition to the personal protective gear specified in point 2.11.3, a protective shield or eyewear shall be used when preparing suspensions of organs, infecting animals, and working with blood.

2.11.5. Work clothes must be changed as they become soiled, but at least once each week.

2.11.6. Before being sent out for laundering, protective clothing must be decontaminated.

2.11.7. Workers who trap rodents or collect arthropods and perform other field work involving wild vertebrate animals and arthropods must be provided with clothing that is appropriate for the season and provides protection.

2.12. Requirements regarding disinfecting various objects and cleaning spaces --Means and methods

As per Russian Federation Chief Health Inspector Decree No. 42, dated 2 June 2009, changes taking effect on 1 August 2009 shall be inserted in point 2.12.1 of this attachment.

See the text of this point in the previous edition.

2.12.1. During work involving group 3 and 4 pathogenic biological agents, the various objects shall be disinfected by using physical (boiling, wet saturated steam under excess pressure, dry hot air, or ultraviolet radiation), chemical (use of disinfectant solutions), and biological (biological ovicides) methods.

The only disinfectants and disinfestants (biological ovicides) and equipment (disinfection chambers, steam and air sterilizers, sprayer devices and units, bactericidal irradiation machines, washing machines, bacterial filters, sterilization boxes, and so forth) that shall be permitted for use in disinfection are those that have been duly authorized for use in industrial manufacture and for use in the Russian Federation.

Disinfection methods and means shall be determined on a case-by-case basis depending on

the pathogenic biological agent and the nature of the material being decontaminated.

2.12.2. When disinfection is performed, preference shall be given to the physical method because of its reliability and safety for personnel.

As per Russian Federation Chief Health Inspector Decree No. 42, dated 2 June 2009, point 2.12.3 of this attachment shall be stated in a revision that will take effect on 1 August 2009.

See the text of this point in the previous edition.

2.12.3. Disinfection using the physical method shall be performed by

- The steam method (in a steam sterilizer);
- The air method (in an air sterilizer);
- The steam and air method (in a disinfection chamber);
- Ultraviolet radiation; and
- Microwave currents (for wastes).

Boiling in a disinfecting boiler in water or in water to which bicarbonate of soda (baking soda) has been added shall also be permitted.

2.12.4. Disinfection by the boiling method shall be used for vessels (including laboratory vessels), linens, personnel's protective clothing, rubber gloves, rubber hoses, stoppers, pipette bulbs for pipetting infected material, instruments that have been used to dissect laboratory animals, liquid waste, wash water, stoppers, cleaning supplies, bags for transporting wild rodents, and so forth.

2.12.5. The steam method shall be used to disinfect laboratory vessels, personnel's protective clothing, bacteriological cultures, containers and bowls for animals, bedding material, animal excrement, leftover feed, metal cages, bowls in which animals were dissected and implements used in trapping, bacterial air filters, animal carcasses, liquid wastes, and wash water.

2.12.6. The air method shall be used to disinfect laboratory vessels made of glass, metals, or loose silicone rubber. This method shall be used to disinfect glassware that has not been contaminated with organic matter.

2.12.7. The air and steam method shall be used in disinfection chambers to disinfect cotton coats, trousers, bedding, short fur coats, caps, leather and fur footwear, and slippers.

2.12.8. Disinfectants shall be used to decontaminate [the following]: limited sections of soil; surfaces in rooms; furniture; equipment; personnel's protective clothing; linens; rubber gloves; eyewear; footwear; laboratory vessels (pipettes, test tubes, flasks, Petri dishes, glass slides, combs, for drying cultures, syringes, and so forth); instruments (including after laboratory animals have been dissected); metal chests; cages; bowls in which animals have been dissected and implements used in trapping; bedding; liquid wastes; discharge from an ill animal (sputum, urine, and feces); vessels used to collect discharge from an ill animal; sanitation equipment; cleaning equipment; waste bins; and vehicles.

Agents containing the following active ingredients shall be used for disinfection: active oxygen (peroxide compounds and so forth); cationic surfactants; active chlorine compounds; aldehydes; and alcohols (ethanol, propanol, and so forth). They are used most often in the form of multicomponent formulations containing one or more disinfectants and functional additives (anticorrosive, deodorizing, detergent, and so forth).

The procedures for disinfecting different objects that have been contaminated with pathogens belonging to pathogenicity groups 3 and 4 (bacteria [including mycobacteria], viruses, fungi, and bacilli spores) by using disinfectants shall be determined by the specifics of the directions for their use.

2.12.9. The choice of disinfectant shall be determined by the specifics of the objects to be decontaminated and the ultimate purpose of the agent.

During regular and general cleaning involving the use of disinfectant solutions, surfaces in rooms, instruments, equipment, and so forth shall be disinfected by wiping with a fabric cloth or rag that has been moistened with disinfectant solution. For these purposes, it is advisable to use disinfectants with a detergent effect. When emergency treatment of surfaces that have a small area and are hard to reach becomes necessary during the course of a workday, one can use ready-to-use forms of disinfectants (for example, alcohol-based [disinfectants], which are characterized by a short exposure time). This can be done by using hand sprayers, wiping with disinfectant solutions applied to a rag, or using ready-to-use disinfectant cloths. Using a disinfectant with detergent properties makes it possible to combine decontamination of an object with cleaning it. Disinfectants with a detergent effect shall therefore used be for regular and general cleaning.

Active chlorine compounds shall be used mainly for disinfecting discharges (feces, sputum, and so forth) and vessels that were used to collect discharges.

Agents that do not contain aldehydes or alcohols shall be used for disinfecting tableware, protective clothing, and linens.

Agents based on aldehydes, cationic surfactants, hydrogen peroxides, alcohols, and chlorine-containing agents shall be used to disinfect medical products and laboratory vessels. The products and vessels shall be disinfected by submerging them in disinfectant solution. Products that can be disassembled shall be disinfected after they have been taken apart. Products' channels and cavities shall be filled with disinfectant solution.

Agents based on aldehydes, cationic surfactants, hydrogen peroxides, and chlorine-containing agents shall be used to disinfect metal chests, cages, bowls in which animals were dissected, and trapping implements. They shall be disinfected by wiping in accordance with the procedures recommended for decontaminated surfaces or by submersion in accordance with the procedures recommended for decontaminating medical products in the instructions for using the agents.

2.12.10. Before being disposed of, class B and V [*Translator's note*: the second and third letters in the Cyrillic alphabet] medical wastes (linens, masks, protective clothing, cloths, disposable medical problems, and so forth) shall be deco ntaminated at the site where they were generated by submerging them in disinfectant solutions in accordance with the sanitary regulations and standards "Rules for Collecting, Storing, and Removing Wastes From Health Care Institutions." Medical wastes shall be disinfected by using chemical and physical decontamination methods in accordance with procedures guaranteed to kill the respective pathogens. Discharges, blood, sputum, and so forth shall also be disinfected by using dry

active chlorine disinfectants (hypochlorite of lime, neutral calcium hypochlorite, and so forth). Medical wastes may be decontaminated and disposed of simultaneously by using units authorized for use in accordance with the established procedure.

2.12.11. A minimum of a week's supply of disinfectants must be stored in the laboratory.

2.12.12. New lots of disinfectant that enter the warehouse need to be inspected for their content of active ingredient.

2.12.13. Disinfectant solutions shall be prepared in specially designated spaces or an exhaust fume hood. The container of disinfectant solution must have a label indicating its name, concentration, and the date when it was prepared.

2.12.14. Autoclaving shall be performed by personnel who have a certificate of completion of special courses.

The operation of steam and air sterilizers used to decontaminate materials shall be inspected in accordance with the existing instructional-directive and methodological documents by physical, chemical, and biological methods.

A bacteriological inspection of the sterilizers shall be performed after installation and repair of the equipment, as well as during the process of its operation (scheduled twice per year and also when unsatisfactory inspection results are obtained).

2.12.15. Transfer of material for decontamination within the subunit shall be done in special containers (tanks, buckets, and dressing boxes with covers).

2.12.16. Regular cleaning of premises shall be performed weekly by the wet method after the workday has ended: Detergents shall be used in the laboratory's "clean" zone, and disinfectants shall be used in the "contaminated" zone. Disinfection of objects that are contaminated with blood or other biological substrates associated with a risk of spread of parenteral hepatitis viruses and HIV infection shall be guided by the existing instructional and methodological documents, and the disinfection shall be used in an antiviral regimen.

In cubicle spaces, general cleaning of spaces with disinfectants shall be performed weekly. The surfaces in spaces, devices, and instruments shall be wiped with disinfectant solution, and the walls shall be cleaned at a height up to 2 meters. After wet cleaning, bactericidal lamps shall be turned on. Bactericidal irradiation machines shall be used in accordance with the existing duly approved methodological documents on use of bactericidal lamps to disinfect the air and surfaces in premises.

The glass surfaces of bactericidal lamps shall be wiped with a rag that has been moistened with alcohol at least once per week while the lamp is in the off position.

2.12.17. Cleaning supplies for the "clean" and "contaminated" zones must be labeled separately. Moving them from one zone to the other shall not be permitted.

2.12.18. After work has been completed, medical personnel must treat their hands with a disinfectant solution or 70% alcohol and then wash them with soap. Use of skin antiseptics (according to the directions for their use) shall be permitted.

As per Russian Federation Chief Health Inspector Decree No. 42, dated 2 June 2009,

changes taking effect on 1 August 2009 shall be inserted in Section III of this attachment.

See the text of this point in the previous edition.

III. Requirements regarding the procedure for eliminating accidents during work with pathogenic biological agents

3.1. To prepare for an accident resulting in the real or potential possibility of a pathogenic biological agent being released into the air or the human habitat and infecting personnel, subunits that work with pathogenic biological agents must have an accident elimination plan and a stockpile of disinfectants that are active against the pathogens with which tests are being performed.

A subunit that works with pathogenic biological agents must store [the following items] in a specially allocated location: a spray gun (automax); sets of work clothes (into which the affected individuals can change) and protective clothing (for the employees who are eliminating the accident); and an emergency first aid kit.

The emergency first aid kit shall contain the following: 70% ethyl alcohol (two 100-mL bottles); two or three weighed portions of potassium permanganate for preparing a 0.05% solution (0.0125 g potassium permanganate plus 25 mL water); sterile distilled water; 5% tincture of iodine; scissors with curved jaws; dressing materials (absorbent cotton, bandages, and so forth); cord; and ammonia spirit.

In addition to the aforesaid [items], the emergency medical kit of a virology laboratory must also contain 1% boric acid solution and interferon or an interferon inducer, and the emergency kit of a mycology laboratory must also contain 1% boric acid solution or weighed portions for preparing a solution (0.25 g boric acid plus 25 mL water).

Laboratories of scientific research institutes that perform tests on microorganisms in pathogenicity groups 3 and 4 with altered properties must have a stockpile of agents for emergency prevention and treatment (antibiotics, sera, immunoglobulins, and so forth) for two to four individuals.

The head of the subunit shall be responsible for assembling the emergency first aid kit.

3.2. The extent of the measures implemented to eliminate an accident shall depend on the nature of the work that is being performed, the type of pathogen and its properties, and the scale of the accident:

- Accidents involving dissemination of pathogenic biological agents, which is to say formation of an aerosol (breakage of test tubes, bottles, or flasks containing a liquid culture; breakage of dishes and test tubes containing cultures on agar with a condensate; spattering of a bacterial suspension from a pipette or syringe; spraying of tissue fluid when carcasses of infected animals or sick humans are being dissected; accidents in a vacuum unit during the process of drying virulent cultures; accidents leading to contamination of the air or environmental objects; and accidents occurring while pathogenic biological agents are being transported to the autoclave area or between subdepartments);
- Accidents not involving dissemination of a pathogenic biological agent (loops with infected material coming into contact with the edge of a dish, test tube, bottle, or

crystallizer; a crack in a Petri dish, test tube, or bottle containing biological material; a solid particle falling onto a table when a loop is being heated over a flame after an inoculation; touching the surface of a culture on solid culture medium; and so forth); and

- Accidents associated with interruption of the skin's integrity.

3.3. Employee actions during an accident:

3.3.1. In the event of an accident involving dissemination of a pathogenic biological agent

- All individuals who are in the space shall immediately stop working. While holding their breath, they shall exit the contaminated space and move into the dressing area, after which they shall close the door tightly, activate the emergency alarm system, and notify the subunit's head of what has occurred;
- Hands shall be treated with disinfectant solution or alcohol, and if the individual was not been protected, he shall be treated liberally with 70% ethyl alcohol;
- The mucous membranes, nose, and mouth shall be treated with preparations from the emergency first aid kit: The mouth and throat sh all be rinsed with 70% ethyl alcohol, drops of a 1:100,000 solution of potassium permanganate or 1% boric acid solution shall be placed in the nose, and (in the case of an accident involving viruses), drops of interferon or an interferon inducer shall then be administered;
- The protective clothing shall be removed and submerged in disinfectant solution or placed in a dressing box (receptacle) for autoclaving;
- Exposed parts of the body shall be rubbed with 70% ethyl alcohol;
- Solutions of antibiotics or other agents to which the pathogen is susceptible shall be administered in drops into the eyes (and possibly into the nose);
- A cleansing shower shall be taken; and
- Clean work clothing shall be put on.

Procedure for conducting disinfection:

- The employees who are taking part in eliminating the accident must be dressed in hazmat suits (surgical scrubs), head scarfs, and overshoes (plastic surgical boots);
- During disinfection by the spraying method, type RU-60 M or RPG-68 respirators with a cartridge appropriate for the disinfectant being used or a type GP-5 gas mask must be used as personal protective gear for the respiratory organs;
- A disinfectant solution that is effective in relation to the infectious agent in question shall be used for the treatment;
- The space shall be disinfected by using a spray gun (automax) to spray disinfectant solution from the entrance door onward, moving along the treated area and spraying all objects in front of oneself (the floor, walls, and ceiling) and into the air;
- Two hours after the initial treatment, the cotton plugs moistened with disinfectant solution and fragments of broken glassware shall be collected and submerged into a container holding disinfectant solution, and laboratory vessels containing cultures that were located on working surfaces at the time of the accident shall be submerged into a container holding disinfectant solution or wiped that has been with a cloth moistened with disinfectant solution and placed in a container for autoclaving;
- After the disinfection has been completed, the air and surfaces in the space shall be disinfected with bactericidal lamps (using procedures in accordance with the normative documents);

- The employee who performed the disinfection shall exit the dressing area or corridor, remove his protective clothing, and submerge it in disinfectant solution; and
- After 2 hours, the space shall be cleaned, after which work may be resumed.

3.3.2. In the event of an accident not involving dissemination of a pathogenic biological agent

- Without leaving the space, [the employee] shall place a cotton plug with disinfectant solution on the location of site on the object's surface that has been contaminated with a pathogenic biological agent;
- The emergency alarm system shall be activated, the subunit's head or his surrogate shall be called, and disinfection treatment of the accident site shall be continued; and
- After the disinfection treatment has been completed, the employee shall exit the space where the accident occurred, and he shall remove his protective clothing and submerge it in disinfectant solution; and
- Exposed parts of his body shall be treated with disinfectant solution or 70% alcohol.

3.3.3. In the event of an accident involving interruption of the skin's integrity:

- Work shall be halted;
- The emergency alarm system shall be activated;
- Hands shall be treated with a disinfectant solution, gloves shall be removed, and blood shall be squeezed from the wound and into the disinfectant solution;
- A compress consisting of disinfectant solution or 70% ethyl alcohol shall be placed on the wound for 4 to 5 minutes; and
- During work with viruses, blood shall be squeezed out onto a dry sterile pad, and the wound shall be treated with a 5% tincture of iodine without the use of a disinfectant solution.

3.3.4. In the event of an accident during work on a centrifuge, the cover shall be opened slowly, but only after at least 30 to 40 minutes (after the aerosol has settled). The centrifuge bags and broken glass shall be placed in a disinfectant solution, and the surface of the centrifuge's cover, interior areas, and outer surface shall be disinfected. The centrifuge shall be disinfected after it has been disconnected from the electrical power supply.

3.4. After an "accident" alarm, any employee who has heard the signal shall quickly inform the subunit's head (or a specialist who is his surrogate) of what has occurred.

The subunit's head shall report the accident to the committee for monitoring compliance with biological safety requirements and to the organization's head.

3.5. The head of the subunit and chairman of the committee for monitoring compliance with biological safety requirements shall assess the situation, determine the extent of the measures to localize the accident and mitigate its effects, and report to the organization's head. They shall also organize and monitor the actions of the employees involved in eliminating the accident.

3.6. The laboratory's head shall submit a written report about the accident that occurred and

measures implemented to the organization's head and to the chairman of the committee for monitoring compliance with biological safety requirements. It shall indicate the time and date of the accident that occurred, its nature, the employees who were located at the accident site (including those who implemented the disinfection measures), and the measures that were implemented.

3.7. After the accident has been contained, the organization's head and members of the committee for monitoring compliance with biological safety requirements shall jointly assess the scope and quality (including involving the use of laboratory testing methods) of the steps that were taken to contain or eliminate the accident and make the decision regarding resuming work with microorganisms.

3.8. In all subunits that work with pathogenic biological agents, scheduled training exercises dealing with responding to accidents must be held at least once per year.

IV. Organization of monitoring of compliance with biological safety requirements

As per Russian Federation Chief Health Inspector Decree No. 42, dated 2 June 2009, changes taking effect on 1 August 2009 shall be inserted in point 4.1 of this attachment.

See the text of this point in the previous edition.

4.1. Public health and epidemiological surveillance of compliance with the requirements of these regulations in subunits working with pathogenic biological agents shall be conducted by the regional offices of the Federal Service for Surveillance in the Area of Protection of Consumer Rights and Human Welfare, institutions and the structural subunits of the federal authorities that conduct public health and epidemiological surveillance at defense and other special facilities.

4.2. A committee to monitor compliance with biological safety requirements shall be created at organizations that work with pathogenic biological agents.

4.3. Day-to-day monitoring of compliance with the requirements of these regulations shall be conducted by the laboratory head or by an individual designated by an order concerning the organization.