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Manual for biological experiments (second edition)

—For SPring-8/SACLA users



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1. Introduction

This textbook provides, in order to ensure the safety of research conducted at SPring-8/SACLA, health and safety knowledge and the rules for the facility users engaged in biological experiments. In biological experiments, various biological materials are used. Before drafting a research/experiment plan, you must understand all the properties of the biological material you are going to use and learn how to use the devices and the like prior to starting an experiment.

Biological experiments, including genetic recombinant experiments, animal experiments, experiments involving microorganisms and research involving human subjects (human derived samples/information), must conform to the laws, regulations, guidelines, and the rules specified by SPring-8/SACLA. Please note that you must observe these rules to ensure the safety of workers engaged in such experiments, other researchers and the general community.

Research involving human subjects (safety matters conform to those of microorganism experiments) and animal experiments should be conducted with full consideration given to research ethics and animal welfare respectively.

2. Precautions for handling biomaterials

To conduct research involving biomaterials in a safe manner, observe the following two major points:

- (1) Consider risks of damage to the environment, including beyond the location at which the experiment is performed, and risks to living organisms, including workers engaged in experiments, caused by microorganisms and biologically active agents contained in experimental biomaterials.
- (2) Use experimental equipment and tools in a proper manner.

2-1. Infection, allergies and poisoning due to biological materials

(1) Infection by biomaterials

Handling microorganisms and experimental animals involves the risk of infection. Always wear appropriate protective gear such as a laboratory coat, rubber gloves, protective glasses and a mask to prevent infection. In order to avoid secondary infection, inactivate fungus bodies and culture fluid using an autoclave and the like before disposal.

(2) Allergies due to biomaterials

When handling materials that contain allergens, use mechanical pipettes and wear protective gear including a laboratory coat, rubber gloves and protective glasses to avoid direct contact with the materials. Waste that contains allergens must be put in a sealed plastic bag and the like to be disposed of safely.

Handle experimental animals wearing appropriate protective gear because experimental animal hair, dust from the breeding rooms and animal bites can be allergens.

(3) Poisoning caused by biomaterials

When conducting biological experiments using toxins and the like produced by organisms, handle them, in a similar way to handling dangerous chemicals, carefully and while wearing appropriate protective gear, including rubber gloves.

2-2. Handling biochemical reagents

Some reagents used in biological experiments possess high carcinogenicity, teratogenicity, neurotoxicity, and allergenicity. Wear proper protective gear when using such reagents.

After processing biomaterials containing such substances in an autoclave, wait for them to cool down before removing them, as vapor from items that are taken out of the device shortly after autoclaving may contain inactivated substances.

When handling poisonous and deleterious substances, refer to the following websites:

http://www.spring8.or.jp/ja/users/safety/form_procedure/chemistry (in Japanese)

http://www.spring8.or.jp/en/users/safety/form_procedure/chemistry (in English)

2-3. Handling experimental/analytical devices

In order to use devices in a safe manner, refer to the manuals and, before handling complex devices in experiments, have an experienced person demonstrate how to use it. Confirm that reagents and the like used in a device are set in place.

In biological experiments, the following accidents have been reported: i) burns caused by heating gel with a microwave oven, ii) burns resulting from contact with a hot autoclave, iii) injury of the eyes during UV radiation while not wearing protective glasses for UV rays, and iv) fire/smoke resulting from improper setting of the temperature of a dry heat sterilizer.

Accidents were also caused by improper handling of injection needles, surgery instruments, microtome blades, and other tools with sharp edges. Workers must handle experimental equipment carefully and dispose of it in a proper manner.



When handling microwave ovens, autoclaves, and transilluminators, wear appropriate protective gear.



Be careful to set the temperature of dry sterilizers.



Wear cut-resistant gloves when handling blades.



Handling liquid nitrogen, which is often used for instant freezing of biomaterials, also causes accidents. Examples include frostbites due to contact with liquid nitrogen and rupture of freezing tubes being thawed. When handling liquid nitrogen, wear protective glasses and cryo-protective gloves while taking necessary safety measures. Adequately ventilate the room where liquid nitrogen is being used. Using liquid nitrogen in a closed place can cause suffocation.



Experiments using the following items must conform to related laws and regulations and guidelines, and rules specified by the SPring-8/SACLA: i) dangerous devices that produce high temperature, vapor, high pressure gases, UV rays, high voltage, and laser beams, and ii) labelled compounds containing radioisotopes. When conducting such experiments, observe these laws, rules and the like.

3. Experiments involving microorganisms

Microorganisms, etc.* should be handled in conformity with related laws, regulations, and the rules specified by SPring-8/SACLA. Only level 1 and 2 microorganism experiments are allowed to be conducted on the Harima Campus.

* Microorganisms, etc. refer to bacteria including chlamydia and rickettsia, viruses, viroid,

fungi, parasites and biological materials (including human derived samples) contaminated with them, but exclude genetically modified organisms.

Levels of microorganisms, etc.

Level 1	Have no risks of causing significant infection to humans or animals
Level 2	Pathogenic to humans and animals but have a low possibility of causing serious danger inside the laboratories, to other workers, and to the environment.

3-1. Required procedures

Before handling microorganisms in research in SPring-8/SACLA, the procedures shown below must be followed:

(1) Applying research plans

When using materials that can damage the human body, livestock, and agricultural and marine products through biological interaction, submit the Biological Experiment Plan and the Safety Instruction Manual to the Users Office of Japan Synchrotron Radiation Research Institute (JASRI) by the designated date. Materials that require submission include: pathogenic microorganisms such as bacteria, viruses, fungi, and infectious nucleic acids/plasmids; parasites and toxic substances produced by them; carcinogens, allergens, and other similar materials."For details, refer to the following websites:

http://www.spring8.or.jp/ja/users/safety/form_procedure/biology (in Japanese)

http://www.spring8.or.jp/en/users/safety/form_procedure/biology (in English)

When handling human derived samples, the procedures for an experiment involving human subjects must also be followed, in addition to the procedures above. See 6-1. Required procedures in 6. Research involving human subjects.

(2) Handling legally regulated microorganisms, etc.

Before handling i) export prohibited items specified by the Plant Protection Act, ii) specific pathogens designated by the Act on Prevention of Infectious Diseases and Medical Care for Patients Suffering Infectious Diseases, iii) pathogens specified by the Act on Domestic Animal Infectious Diseases Control, and iv) microorganisms regulated by other laws and regulations, you must submit an application for approval or a notification to the government authorities. When you are planning to use such items, consult JASRI Safety Office as early as possible.

(3) Carrying in microorganisms

a) Level 1 microorganisms

Submit the List of Samples, Reagents, etc. via my page on the User Information page of SPring-8/SACLA no later than 10 days before each visit.

<https://user.spring8.or.jp/apps/login/en?institution=0> (in English)

b) Level 2 microorganisms

Submit a written Transport of Infectious Agents Designated Biosafety Level 2 to JASRI Users Office no later than two weeks prior to the date of carry-in. For details, refer to the following websites:

SPring-8: <https://user.spring8.or.jp/?p=1565> (in Japanese)

SACLA: <http://sacla.xfel.jp/?p=89> (in Japanese)

SPring-8: <https://user.spring8.or.jp/?p=1565&lang=en> (in English)

SACLA: <http://sacla.xfel.jp/?p=89&lang=en> (in English)

Submit to: JASRI Users Office, tel: 0791-58-0961, email: sp8jasri@spring8.or.jp

3-2. Precautions for conducting experiments

(1) Location of experiments

Microorganisms must only be handled in approved experimental rooms, where drinking, eating and storing food are prohibited.

(2) Signs

Post an international biohazard sign and a label indicating the level of the microorganisms at the entrance to the laboratories and storage cabinets where microorganisms are actually handled and stored.



Post the signs at the entrance.

(3) Containment measures/infection preventive measures

When conducting experiments using microorganisms, observe the following rules to prevent the spread of contamination, damage to the environment, and infection accidents.

Note that human derived samples are classified into level 2 and must be handled as infectious material. When handling infectious viruses such as hepatitis B viruses, you must adequately consider i) equipment, ii) methods, iii) incubation period, and iv) the possibility that other infectious known and unknown viruses are present.

- 1) Prepare disinfectants and sterilizers appropriate for the microorganisms you use.
- 2) Avoid wearing clothes, sandals and the like that allow skin to be exposed.
- 3) Wear appropriate protective gear such as a laboratory coat, rubber gloves, protective glasses, and a mask.
- 4) Close the windows and doors of the experimental room to prohibit entry of non-authorized persons.
- 5) When handling microorganisms, the use of a biohazard cabinet is recommended.
- 6) Keep the location of experiments in order.
- 7) Disinfect and sterilize inside of equipment and tools, and wash your hands when necessary.
- 8) When taking microorganisms out of an experimental room, put them in a container that prevents them from leaking and spreading.
- 9) When using microorganisms in the experimental hall (including the experimental hutch), take necessary measures to avoid scattering of them. In the experimental hutch, where their leakage may be caused by beam radiation, take necessary measures to avoid scattering of them.
- 10) Store solid/liquid waste in a cool dark place after disinfecting/sterilizing or sealing up.

When handling level 2 microorganisms, also take the following measures:

- 11) Explain to others working in the vicinity all of the hazardous properties of the microorganisms and the like that you use prior to starting an experiment.
- 12) When handling microorganisms, take measures to limit aerosols to the least amount possible. When aerosols are generated or when there is such a possibility, conduct experiments in a biohazard cabinet.

(4) Storage

Levels 1 and 2 microorganisms should be stored using dedicated storage containers in approved experimental rooms. If such an experimental room is vacant, lock the storage cabinets or the room.

When they are stored in a sealed container that can avoid leakage of the contents, or when other similar measures are taken, they can be stored in a storage cabinet outside of the experimental room where they are actually used.

(5) Disposal

Disinfect and sterilize waste originating from your experiment and bring it back to your laboratory to dispose of.

Limit the items that can cause infection to the least amount possible and disinfect/sterilize them totally before disposal to prevent contamination and spreading.

Disinfect/sterilize used biomaterials such as bacterial cells/culture fluid and disposable experimental tools that are stained with such materials using autoclaves and the like before disposal.

Before washing laboratory ware, always disinfect and sterilize it. The primary and secondary rinse water should be collected as waste liquid.

4. Genetic recombinant experiments

They must be conducted in conformity with related laws and regulations and the rules specified by SPring-8/SACLA.

On the Harima campus, only the following experiments are allowed to be conducted: i) experiments that require P1 and P2 level containment measures, ii) P1A and P2A animal experiments and iii) P1P and P2P experiments using plants.

4-1. Required procedures

When conducting genetic recombinant experiments at SPring-8/SACLA, the following procedures are required:

(1) Applying research plans

When conducting genetic recombinant experiments at SPring-8/SACLA, apply a research plan and undergo the committee's examination process to obtain approval. Submit the Application for Approval of Genetic Recombination Experiment to JASRI Users Office by the designated date.

For details, refer to the following websites:

http://www.spring8.or.jp/ja/users/safety/form_procedure/biology (in Japanese)

http://www.spring8.or.jp/en/users/safety/form_procedure/biology (in English)

When handling animals (mammals, birds, and reptiles), submit the Animal Experiment Application Form to JASRI Users Office, separately from obtaining approval of the Application for Approval of Genetic Recombination Experiment, to have it examined by the committee for

approval as an animal experiment. See 5-1. Required procedures in 5. Animal experiments. For more details, refer to the following websites:

http://www.spring8.or.jp/ja/users/safety/form_procedure/biology (in Japanese)

http://www.spring8.or.jp/en/users/safety/form_procedure/biology (in English)

(2) Carrying in/out genetically modified organisms

Submit a written Genetically Modified Organisms Carry In/Out Notification Form to JASRI Users Office at least ten days prior to the date of work. For more details, refer to the following websites:

http://www.spring8.or.jp/ja/users/safety/form_procedure/biology (in Japanese)

http://www.spring8.or.jp/en/users/safety/form_procedure/biology (in English)

Submit documents to: JASRI Users Office, tel: 0791-58-0961, email: sp8jasri@spring8.or.jp

4-2. Precautions for conducting experiments

(1) Location of experiments

Genetic recombinant experiments must be conducted in approved experimental rooms with the doors closed. In order to prevent persons who are not engaged in the experiments from entering the room without permission, post a sign indicating authorized personnel only at the entrance to the room, lock the room when it is vacant and take other necessary security measures.

Experiments using genetically modified animals/plants must be carried out in a location that is designed to prevent such animals and pollen of such plants from escaping and scattering.

Drinking, eating and storing foods are prohibited in experimental rooms.

(2) Signs

For P2, P1A, P2A, P1P and P2P level experiments including culturing, breeding, and raising genetically modified organisms, the signs shown in table 1 are required to be posted, depending on the content of experiments.

When storing genetically modified organisms intended for use in experiments in an experimental room temporarily, which is legally considered as “experiment underway”, take the same measures. The signs must be also posted at the storage facilities.

When a sign indicating experiment underway is posted, close the door of the experimental room. If you leave the door open while experiments are not being conducted in the room, remove or cover the signs, and take other measures to make them invisible.

Table 1 Required signs and location to post

Level	Sign	Location to post
P1A	<div style="border: 1px solid black; padding: 5px; text-align: center;">組換え動物等飼育中</div> Genetically modified animals now breeding	Entrance to experimental room
P1P	<div style="border: 1px solid black; padding: 5px; text-align: center;">組換え植物等栽培中</div> Genetically modified plants now raising	Entrance to experimental room
P2	<div style="border: 1px solid black; padding: 5px; text-align: center;">P2レベル実験中</div> P2 level experiment going on	Entrance to experimental room and storage equipment
P2A	<div style="border: 1px solid black; padding: 5px; text-align: center;">組換え動物等飼育中</div> Genetically modified plants now raising	Entrance to experimental room
P2P	<div style="border: 1px solid black; padding: 5px; text-align: center;">組換え植物等栽培中</div> Genetically modified plants now raising	Entrance to experimental room



Example of posting signs at the entrance to P1A experimental room (genetic recombinant experiments)

(3) Containment measures

When handling genetically modified organisms in SPring-8/SACLA, which falls under a category of Type 2 Use under the laws, take containment measures that are specified by the laws and regulations or approved by the Minister of Education, Culture, Sports, Science and Technology. Please observe the following rules:

- 1) Prepare agents and sterilizers that can adequately inactivate the genetically modified

organisms you use.

- 2) Avoid wearing clothes, sandals and the like that allow skin to be exposed.
- 3) Wear appropriate protective gear such as a laboratory coat, rubber gloves, protective glasses, and a mask.
- 4) Close the windows and doors of experimental rooms where the experiments are being conducted.
- 5) When handling genetically modified animals, take appropriate escape-prevention measures and containment measures depending on each animal species (for example, rodent barriers).
- 6) When performing an operation that can generate aerosols and scatter genetically modified organisms and the like, use tools, devices, and equipment that prevent biohazard.
- 7) Operations in P2 level experiments that can generate aerosols should be conducted in a biohazard safety cabinet.
- 8) Keep the location of experiments in order.
- 9) Disinfect and sterilize inside of the equipment and tools, and wash your hands when necessary.
- 10) When equipment (including a lab bench), devices, and tools are contaminated with genetically modified organisms, or when there is such a possibility, disinfect and sterilize these items after experiments.
- 11) When transferring genetically modified organisms, put them in a container that can prevent them from leaking, escaping, and scattering.
- 12) When using genetically modified organisms in the experimental hall (including the experimental hutch), take necessary measures to avoid scattering of them. In the experimental hutch, where their leakage may be caused by beam radiation, take necessary measures to avoid scattering of them.
- 13) Store solid/liquid waste in a cool dark place after disinfecting/sterilizing or sealing up.

(4) Storage during experiments

Genetically modified organisms must be stored in an approved experimental room using containers that can prevent them from leaking, escaping and scattering. Post a sign indicating that P2 experiments are underway at the storage equipment where P2 level genetically modified organisms are actually stored.

(5) Disposal

Disinfect and sterilize waste and the like originating from your experiment and bring it back to your laboratory to dispose of.

Before disposing of genetically modified organisms, biomaterial before genetic recombinant, and

tools contaminated with such material, always disinfect and sterilize them by autoclaving and the like.

Before washing laboratory ware, always disinfect and sterilize it. The primary and secondary rinse water should be collected as waste liquid.

5. Animal experiments

They must conform to the related laws and regulations and the rules specified by SPring-8/SACLA.

Before conducting animal experiments, serious consideration must be given to i) purpose and necessity of animal use, ii) prevention of unnecessary repetition, iii) search for alternative methods, iv) limiting animals to use to the least number possible, v) pain intensity felt by experimental animals and appropriate methods to alleviate their pain. You must also improve your experimental techniques and give full consideration to animal welfare based on 3R principles (Replacement, Reduction, Refinement).

5-1. Required Procedures

When conducting animal experiments at SPring-8/SACLA, the following procedures are required:

(1) Applying research plans

When conducting animal experiments at SPring-8/SACLA, apply a research plan and undergo the committee's examination process to obtain approval. Submit the Animal Experiment Application Form to JASRI Users Office by the designated date. For details, refer to the following websites:

http://www.spring8.or.jp/ja/users/safety/form_procedure/biology (in Japanese)

http://www.spring8.or.jp/en/users/safety/form_procedure/biology (in English)

When handling genetically modified animals and inoculating animals with genetically modified organisms in experiments, in addition to obtaining approval with the Animal Experiment Application Form, submit the Application for Approval of Genetic Recombination Experiment to JASRI Users Office and undergo the committee's examination to obtain approval as a genetic recombinant experiment. When conducting infection experiments using microorganisms that are not genetically modified, similarly, submit the Biological Experiment Plan and the Safety Instruction Manual for an experiment involving microorganisms as well as the Animal Experiment Application Form for an animal experiment to JASRI Users Office and have them examined by the committee to obtain approval. Refer to 4-1. Required procedures of 4. Genetic Recombinant experiments and 3-1. Required procedures of 3. Experiments involving microorganisms. For details, refer to the following websites:

http://www.spring8.or.jp/ja/users/safety/form_procedure/biology (in Japanese)

http://www.spring8.or.jp/en/users/safety/form_procedure/biology (in English)

Before handling narcotics, stimulants, and psychotropics, you should apply for licenses or identification certificates to the government authorities. When you are going to handle any of such substances, consult JSARI Safety Office as early as possible. For details, see the following websites:

http://www.spring8.or.jp/ja/users/safety/form_procedure/chemistry (in Japanese)

http://www.spring8.or.jp/en/users/safety/form_procedure/chemistry (in English)

(2) Carrying in experimental animals

When delivering experimental animals from outside to SPring-8, submit a “notification of delivery” at least one week before the delivery date.

For details, refer to the following websites:

http://www.spring8.or.jp/ext/ja/bio_formmail/sfm_form_en.html (in English)

When importing experimental animals, before bringing them into SPring-8/SACLA, the prescribed procedures are required by the animal import/export notification system, which conforms to the Act on Prevention of Infectious Diseases and Medical Care for Patients Suffering Infectious Diseases. When you are planning such action, contact JASRI Safety Office as early as possible.

Submit Documents to: JASRI Users Office, tel: 0791-58-0961, e-mail : uoffice@spring8.or.jp

5-2. Precautions for conducting experiments

(1) Location of experiments

Use of experimental animals, that includes experiments, storage, and breeding, must be conducted in approved experimental places. Never eat, drink, and store foods in such places.

(2) Signs

Some animal species such as macaques need legally required signs to be posted.

特 定 動 物	
この動物は人の生命、身体及び財産に害を加えるおそれがある動物であるため、第三者の接触等を禁止します。	
許 可 年 月 日	平成19年5月16日
有効期間の末日	平成24年5月15日
許 可 番 号	第2-34号
特定動物の種類	ニホンザル

Sign indicating specific animals (dangerous animals) now being bred and prohibiting entry of non-authorized persons

(3) Escape/infection/allergy-prevention measures

When handling experimental animals, observe the following rules:

- 1) Prepare disinfectants and sterilizers appropriate for the experimental animals you use.
- 2) Conduct an inspection to check if experimental animals are infected with pathogenic microorganisms when necessary.
- 3) Monitor your health condition every day and avoid contact with experimental animals when you do not feel well.
- 4) Avoid wearing clothes, sandals and the like that allow skin to be exposed.
- 5) Wear appropriate protective gear such as a laboratory coat, rubber gloves, protective glasses, and a mask.
- 6) In order to prevent experimental animals from escaping and scattering into the natural environment, take appropriate preventive measures such as placing rodent barriers, closing the doors of experimental rooms and preparing capture tools.



Rodent barrier placed at the door



Capture net

- 7) Use local exhaust ventilations such as biohazard cabinets when necessary.
- 8) Keep the location of experiments in order.
- 9) Disinfect and sterilize inside of the equipment and tools, and wash your hands when

necessary.

- 10) Transfer experimental animals using transport containers appropriate for animal species. (for example: put animals in a carry bag as well as their cage, use a cardboard box with wire mesh or transport container used by experimental animal distributors.)



Transport box for experimental animals



Cardboard box for transporting experimental animals

- 11) When handling experimental animals in the experimental hall (including the experimental hutch), take necessary measures to avoid animal escape.
- 12) Store solid/liquid waste in a cool dark place after disinfecting/sterilizing or sealing up.

(4) Humane endpoints and euthanasia

In experiments involving a high degree of pain, anesthesia or analgesia must be administered to the animals. When such procedures obstruct the experiments and cannot be used however, set a humane endpoint that can quickly relieve the animals of their pain and achieve the purpose of experiments.

When euthanizing experimental animals, administer an anesthetic to them exceeding its fatal dose or take other appropriate measures to make them unconscious quickly leading to death with the least pain possible. Make sure that the animals are dead.

(5) Experimental operations

In order to ensure animal welfare and the safety of workers engaged in animal experiments, perform proper experimental operations and acclimate experimental animals to a new environment prior to handling them in experiments.

When necessary, anesthetize and restrain them in a proper manner. Effects obtained by anesthesia and restraint depend on the experience of those who actually perform such procedures. Beginners must receive instructions from experienced persons.

When handling narcotics, stimulants, and psychotropics, refer to the website below:

http://www.spring8.or.jp/ja/users/safety/form_procedure/chemistry (in Japanese)

http://www.spring8.or.jp/en/users/safety/form_procedure/chemistry (in English)

Perform the following operations with full consideration given to the matters below:

1) Anesthesia and sedation

Experiment workers should do their best to spare experimental animals' unnecessary pain. Use anesthetics, sedatives or tranquilizers in a proper manner and within the extent necessary for the purpose of the animal experiment.

2) Restraint

When necessary, experimental animals should be restrained in order to relieve the pain they may suffer, to facilitate experimental procedures or to prevent hazards for investigators. But animals should not be restrained any longer than the minimum amount of time necessary. Animals should be acclimated in advance to restraint devices or behavior observation devices.

Prior to restraining an animal, monitor it carefully. If it is agitated, wait until it calms down.

3) Surgery and post-surgical monitoring

Animals should be operated under anesthesia and asepsis; anesthetic monitoring should be maintained during surgery. After surgery, anesthetic recovery and body temperature should be monitored. Prevention of post-surgical infections must be given a high priority. When the animal appears to experience severe pain, appropriate measures should be taken such as administration of a sedative. If necessary, euthanasia (humane endpoint) should be considered.

(6) Monitoring experimental animals

Experiment workers should observe each experimental animal, everyday if possible, from its arrival in the animal facility until the end of the experiment, and give adequate treatment when necessary.

When they spot a dead animal or an animal that is experiencing a health problem, they should notify the principal investigator as well as the animal care manager. Ailing or injured animals should immediately receive adequate treatment or be euthanized.

(7) Procedures at the end of an experiment

When carrying out an experimental animal after an experiment, make sure that the transport container size and the method and time of transferring are appropriate from humane and hygiene perspectives. When the experimental animal cannot be used in another experiment, it should be euthanized.

(8) Disposal of dead animals and the like

Carcasses and the like (including tissues) of experimental animals as well as unsanitary items such as bedding should be appropriately stocked or disposed of, to prevent their impact on the environment and public hygiene.

Carcasses and the like of experimental animals must be kept frozen so that they do not putrefy before their final disposal. Unsanitary items like used bedding should also be stocked appropriately, to prevent their smell and the like from affecting human health and the environment.

Bring back carcasses of experimental animals and unsanitary items such as used bedding to your laboratory to dispose of after disinfecting or sterilizing them when necessary.

Before washing laboratory ware, always disinfect or sterilize it and collect the primary and secondary rinse water as waste liquid.

(9) Care of experimental animals

Experimental animals should be taken care of lawfully and appropriately in compliance with the Standards Relating to the Care and Management of Laboratory Animals and Relief of Pain (Notice No.88 of the Ministry of the Environment).

6. Research involving human subjects

Such research must conform to the related laws and regulations and the rules specified by SPring-8/SACLA.

When conducting the research, make sure that you i) respect the welfare of the donors, ii) respect the intention of the donors to decide whether or not they participate in the research, iii) obtain the informed consent, and iv) observe other ethical codes specified by the Ethical Guidelines for Medical and Health Research Involving Human Subjects (Declaration of Helsinki adopted by the General Assembly of the World Medical Association in 1964).

6-1. Required procedures

When conducting experiments involving human derived samples/information at SPring-8/SACLA, the following procedures are required:

(1) Applying research plans

When conducting an experiment involving human derived samples/information at SPring-8/SACLA, submit the Application Form for the Handling of Human Materials to JASRI Safety Office, no later than two weeks prior to carrying in the items, to obtain approval from the

Safety Office. For details, refer to the following websites:

http://www.spring8.or.jp/ja/users/safety/form_procedure/biology (in Japanese)

http://www.spring8.or.jp/en/users/safety/form_procedure/biology (in English)

Make sure that the experiment has already been approved by the institution with which you are affiliated.

When handling human derived samples that are not identified to be non-infectious, submit the Biological Experiment Plan Safety and the Instruction Manual to JASRI Users Office, separately from obtaining approval of the Application Form for the Handling of Human Materials, and undergo the committee's examination to obtain approval. See 3-1. Required procedures of 3.

Experiments involving microorganisms. For details, refer to the following websites:

http://www.spring8.or.jp/ja/users/safety/form_procedure/biology (in Japanese)

http://www.spring8.or.jp/en/users/safety/form_procedure/biology (in English)

(2) Carrying in/out human derived samples

Submit a List of Samples, Reagents, etc. via my page on the User Information page of SPring-8/SACLA no later than 10 days before each visit.

<https://user.spring8.or.jp/apps/login/en?institution=0> (in English)

Submit documents to: JASRI Users Office, tel: 0791-58-0961, e-mail: uoffice@spring8.or.jp

6-2. Precautions for conducting experiments

(1) Location of experiments, signs, and infection prevention measures

1) Non-infectious human derived samples

They have no requirements for the location of experiments and do not need signs or infection prevention measures.

2) Human derived samples that are not identified to be non-infectious

Using such samples requires the procedures for research involving human subjects as well as those for research involving microorganisms.

a) Location of experiments

Human derived samples that are not identified to be non-infectious must be handled in approved experimental places. Never drink, eat, and store foods in such places.

b) Signs

Post an international biohazard sign and a label indicating level 2 microorganisms at the entrance to the laboratory and on the storage cabinets.

c) Infection prevention measures

Take the following measures to prevent pollution from spreading to the environment and to avoid infection accidents:

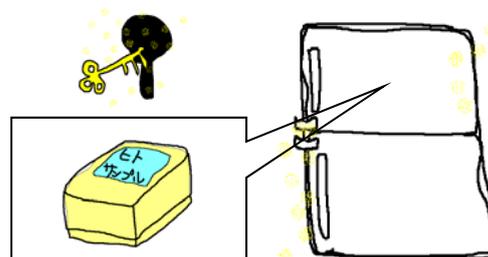
- 1) Prepare appropriate disinfectants and sterilizers.
- 2) Avoid wearing clothes, sandals and the like that allow skin to be exposed.
- 3) Wear appropriate protective gear such as a laboratory coat, rubber gloves, protective glasses, and a mask.
- 4) Close the windows and doors of the experimental room and prohibit entry of non-authorized persons.
- 5) When there are other investigators working nearby, explain to them about the risks of the human derived samples you will be using in advance.
- 6) When handling human derived samples, take measures to limit aerosols produced in the experiment to the least amount possible. Experiments that can generate aerosols must be conducted in a biohazard cabinet.
- 7) Clean and organize the experimental rooms.
- 8) Disinfect and sterilize inside of the equipment and tools, and wash your hands when necessary
- 9) When bringing out human derived samples from the experimental room, put them in a container that can prevent them from leaking and scattering
- 10) When using human derived samples in the experimental hall (including the experimental hutch), take necessary measures to avoid scattering of them. In the experimental hutch, where their leakage may be caused by beam radiation, take necessary measures to avoid scattering of them.
- 11) Store solid/liquid waste in a cool dark place after disinfecting/sterilizing or sealing up.

(2) Anonymization

In SPring-8/SACLA, use anonymized human derived samples and data in principle. When using those that are not anonymized, control them properly to prevent leakage of the personal information.

(3) Storage

Store human derived samples in dedicated storage containers in dedicated storage cabinets,



clearly indicating that the content is such material. If no dedicated cabinets are available, store them clearly separately from other items. Make sure that you lock the storage cabinets and the rooms.

Human derived information must be controlled properly to prevent their theft, loss, leakage and the like.

(4) Waste

Bring back human derived samples to your laboratory after disinfecting or sterilizing when necessary. Never leave human derived information at SPring-8/SACLA.

7. Measures to be taken in case of accidents

(1) In case of minor abnormalities

When an incident occurs that can be handled by personnel working at the site, follow the measures shown below:

- Judge the situation and follow the measures mentioned in (3) to prevent the effects from spreading. But take such action only when your safety is ensured.
- Do not handle the incident alone. Always call someone for help.
- Contact the Beamline Toban (PHS: 3899)

(2) In case of emergency

When discovering leakage and escape of organisms and the like from experimental facilities and/or pollution of the human body and experimental facilities by them, or when noticing a situation potentially leading to such incidents, follow the measures shown below immediately.

- When the damage may spread, evacuate first.
- Contact the Beamline Toban (PHS: 3899)
- If you need to call an ambulance or a fire engine, inform the Guard House (emergency call: 119 or 0791-58-0828) and they will guide the emergency vehicles. The Guard House will alert the personnel and the authorities concerned inside and outside the campus.

(3) When biomaterials are leaked or genetically modified organisms scatter.

When spilling biomaterial, wipe it off to remove. When the material is a genetically modified organism, inactivate the items and the area contaminated, and for infectious biomaterial, disinfect and sterilize those contaminated.

When experimental animals including genetically modified animals escape within the room, capture them immediately while taking measures to prevent them from going out of the room.

When experimental plants including genetically modified plants are spread (seed are scattered), recover them promptly.

(4) First aid for injuries that may cause infection (bites/scratches from experimental animals, contact with pathogenic microorganisms, etc.)

Wash the affected area for 15 minutes or more while squeezing blood from the wound. You can see a nurse at the Health Office (ext: 3299, 999) near Large Door D3 of the Storage Ring Facility between 9:00 and 17:30 on week days.

Immediately contact the Health Office (ext. 3299, 999) when an anaphylactic symptom (serious allergic symptom: hives, breathlessness, chest discomfort, etc.) occurs within half an hour after an injury. During times when a nurse is absent or outside work hours, contact the Guard House (emergency calls: 119, 0791-58-0828) to request an ambulance. Also inform the Guard House of the name of the room/building where the injured person is located and describe the injury, which will be reported to the ambulance on arrival.

Monitor your physical condition carefully for several weeks after an accident to check if such conditions as high fever, rash, lymph node enlargement and overall malaise occur.

When possible, preserve blood serum of the animal that has caused an infection and when the records related to infection (quarantine results) are available, check them.

Manual for biological experiments (second edition)

Emergency calls:

Guard House (ext. 119, 0791-58-0828)

Inquiries:

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