Form20-1 JASRI Safety Office←Person in charge of managing experiments←Project Leader

Safety Supervisor		Confirmation by Safety Office		Receipt by the Safety Office	20YY, MM, DD

Application Form for Genetic Modification Experiment

Date of submission: June 1, 2016

To: The Director General, Japan Synchrotron Radiation Research Institute

(Person in charge of experiment)[1)]
Name of the organization
XX University, Graduate School
Department and title
XX Department XX Assistant
Name (print and signature)
Koukido Hanako

(Manager)[2)]
Name (Leave it blank if User is applying) Seal

To carry out the following genetic modification experiment, I hereby apply for approval of the genetic recombination committee.

Receipt number[3)]	(Do not write when submitting)
Type of application[4)]	☐ New ☐ Renewal (Previous receipt number) ■ Amendment (Previous receipt number Approval 03-10)
Title of experiment[5)]	Examination of metabolism of XXX enzyme in XX deficient mice. The title should concisely describe the purpose and a summary of the experiment
Type of experiment[6)]	■ Microbiology experiment ☐ Large-scale cultivation experiment ■ Animal experiment (■ Animal inoculation・☐ Animal modification) ☐ Plant experiment (☐ Plant inoculation・☐ Plant modification・☐ Fungus modification)
The purpose	The dynamics of XXX enzyme will be examined by infecting mice with a virus that expresses XXX enzyme due to a mutated XX gene.
Summary[7)]	The flow of the relevant genetic modification experiment should be stated.
Expected duration of experiment[8)]	October **, 2016 to March 31, 2019
Contact information of the person in charge of the experiment	Address (Postal code) 1-1-1 Koto, Mikazuki-cho, Sayo-gun, Hyogo Phone (ext./PHS) 07**—**—****(2***) Fax: 07**-**-**** E-mail: hanako@********.**.jp

Prolongation of duration due to the completion of the valid period of the approved experiment → "Renewal"
Change in experimental material or place → "Amendment"
Check appropriate box and write the previous receipt number.

The validity period of the experiment is a maximum of three years from the date of approval. Enter the desirable start date and planned completion date.

Other contacts[9)]	Organization and department of the person in charge of communications XX University, Graduate School XX Faculty XX Lab Office Name of the person in charge of communications Last name First name Address (Postal code) 1-1-1 Koto, Mikazuki-cho, Sayo-gun, Hyogo Phone (ext./PHS) 07**—**—****(3***) Fax: 07**-**-**** E-mail: shikaku@********.**.jp

Place of the experiment (place for animal keeping and raising)[10)] and place for storage of genetically modified organisms[11)]

Building	Room	Containment measures						
		P1	P1A	P1P	P2	P2A	P2P	Storage
Experiment Hall	☐BL20B2 experimental hutch							
	☐BL28B2 optics hutch							
	☐BL40XU experimental hutch							
	☐BL20B2 animal operation room							
	☐Mobile operation room							
	☐BL38B1 experimental hutch							
	☐PXBL experimental hutch							
	☐							
Experimental Animal Facility	■Mouse room		○					
	■Genetic experiment room		○					
	☐Treatment room							
Medium-length Beamline Facility (Experiment building)	☐BL20B2 experimental hutch							
	☐BL20XU experimental hutch							
	☐Animal operation room							
	☐							
Medium-length Beamline Facility (Research building)	☐Room 101							
	☐Room 201							
	☐Room 202							
	☐Room 204							
	☐Room 212							
	☐Room 213							
	■Biochemistry lab 1 Room 208							
	☐Biochemistry lab 2 Room 209							
	☐Biochemistry lab 3 Room 210							
SACLA	☐Experimental hutch (EH3)							
	☐Biological sample prep. room							
	☐							
Others (Write the name of the building and room)	■***** building …… room		○					

Attach documents explaining the followings.
(1) Place of major facility, equipment, and instrument (a detailed drawing of the experimental area
(2 The state of animals or plants that are kept in the room of area, but are not related to the experiment.

Nucleic acid donor/Donor nucleic acid[12)]

Nucleic acid donor	Experiment classification	Donor nucleic acid (type of nucleic acid)	Identification	Note
(Target gene) Streptomyces lavendulae ## strain: genus Streptomyces bacteria of the family Streptomycetaceae	Class 1	XX enzyme gene (cDNA)	Completed/Not completed Completed	

Protostome:Schistosoma japonicum	Class 2	Glutathione S-transferase (cDNA)	Completed	
When changing experimental materials, underline the materials to be added.				
Mouse	Class 1	XX enzyme gene (cDNA)	Completed	
(Expression regulatory gene) E. coli of the family Enterobacteriaceae	Class 1	tac promoter (genome DNA)	Completed	
E. coli of the family Enterobacteriaceae	Class 1	lac promoter (genome DNA)	Completed	
Bacteriophage T7	Class 2	T7 promoter (genome DNA)	Completed	
E. coli of the family Enterobacteriaceae	Class 1	Ampicillin-resistant gene (genome DNA)	Completed	
(Selectable marker gene)				

Host/vector[13)]

Host	Experiment classification	Vector	Type	Note
EK1	Class 1	pUC119 (cloning vector for E. coli) pGEX (vector for protein expression)	Microorganism/Animal/Plant Microorganism	
SD rats and fertilized eggs of SD rats	Class 1	Not used	Animal	

Characteristics of animals, plants, or cells that possess genetically modified organisms.[14)]	Insect cells will be used for cultured cells that will be infected with a genetically modified virus. The cultured cell line that will be used in the experiment will not grow anywhere else than on the artificial medium. The gene inserted into the genetically modified virus will be expressed by infection. The infected cells will die due to virus multiplication. The infected cells will not develop drug resistance.
Table of genetically modified organisms and containment measures.[15)]	as per attached
Method of inactivation of genetically modified organisms.[16)]	Inactivate by autoclaving (121℃, 20 minutes).
Note[17)]	Grant-in-Aid for Scientific Research of Japan Synchrotron Radiation Research Institute will not be used. If public funding (such as Grant in Aid for Scientific Research of the Ministry of Education, Culture, Sports, Science and Technology) is received as Japan Synchrotron Radiation Research Institute, indicate it accordingly.

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※Notes

1) For “Project Leader,” the information shall be given on a person who is directly manage a genetic recombination experiment at SPring-8 and have experience for one year or more. However, a student should not be.
2) For “Person in charge of managing experiments,” the information shall be given on a person who is in charge of administration of this application.
   JASRI, RIKEN, JAERI Staff >> Director
   User >> Director of Users Office （A blank is sufficient in case it submits.）
3) Since "Proposal number" is informed when Safety Office receives this application, leave a column blank. Proposal number is required for all the documents for which you will apply in the future.
4) For “Type of Application,” select any items under which your application falls. In case Continuation or Changes, give the previous proposal number.
5) For “Title,” mention a name that expresses the objective and an outline of a genetic recombinant experiment briefly.
6) For “Type,” select all items under which a genetic recombinant experiment falls.
7) For “Outline,” all living modified organisms involved in a genetic recombinant experiment and the categories of containment measures to be taken during a genetic recombinant experiment shall be mentioned so as to show their processes.
8) The experiment is valid for a maximum of three years from approved day.
9) For “Other contact,” if there is any other contact for administrative matters than Project Leader or Deputy Project Leader, give the information on the contact.
10) For “Laboratory ,Experiment area ,Experiment section (include the area of the breeding animals or culture of plants) ,” select all area under which a genetic recombinant experiment falls. If there is no appropriate column, indicate name of facility and room on proper column and attach the information those mentioned in below.
    ①Names and positions of major facilities, equipment and apparatus;
    ②In case an animal or a plant that has no relation with a genetic recombination experiment is bred or cultured in the laboratory, the experiment section, the experiment area, the breeding section or the screened greenhouse, the state of the breeding of the animal or the culture of the plant;
11） For “Facility of Storage,” select all area that storage living modified organisms in the process of a genetic recombination experiment. If there is no appropriate column, indicate name of facility and room on proper column and attach the information about them.
12) For “Donor organism/Donor nucleic acid,” the following shall be mentioned about the donor organism and donor nucleic acid of the living modified organism for Genetic recombination experiment every component elements (target genes, expression regulatory genes, drug-resistant genes and marker genes).
    a. General name and taxonomical position (familia, genus, species, strain) of donor organism
    b. General name and type (such as genomic nucleic acid, complementary deoxyribonucleic acid or synthesized nucleic acid) of donor nucleic acid.
    c. Attach copy of nucleotide sequence information or an accession number to the nucleotide sequence database of, for example, the Japan DNA Databank (only in the case of donor nucleic acid that is identified nucleic acid).
13) For “Recipient organism/Vector,” the following shall be mentioned about the recipient organism and vector of the living modified organism for Genetic recombination experiment.
    a. General name and taxonomical position (familia, genus, species, strain) of recipient organism
    b. General name, code and short explanation about (ex. pUC119 cloning vector for *E.coli*)
    c For “Type,” select Microorganisms, Animals or Plants.
14) For “Characteristics of animal, plant or cell which retains living modified organisms,” in addition to items those mentioned in below, characters expected to be newly given or already given to an animal, a plant or a cell which retains the living modified organism for Genetic recombination experiment in comparison with animals, plants or cells which do not retain the living modified organisms Genetic recombination experiment shall be mentioned.
    a. Taxonomical position and experiment classification of animal, plant or cell which retains living modified organism;
    b. State of distribution in natural environment and environment in which living or growth is possible;
    c. Pathogenicity, production of harmful substances and other properties;
15) For “Combination Living Modified Organism and its Category of Containment Measures,” all donor organisms, donor nucleic acids, vectors, recipient organisms and animals, plants or cells which retains living modified organism involved in a genetic recombination experiment and the categories of containment measures to be taken during the experiment shall be mentioned so as to show processes of the experiment.
16) For “Measure for inactivating living modified organism,” about the containment measures to be taken during Genetic recombinant experiment, mention a measure for inactivating waste products containing the living modified organism and apparatus and appliances to which the living modified organism sticks, and the effectiveness of the measure.
17) Provide a note when you have received a public grant (such as Grant-in-Aid for Scientific Research from the Ministry of Education, Culture, Sports, Science and Technology) affiliated with JASRI, when the research proposal is for a position paper of the Ministry of Education, Culture, Sports, Science and Technology, or when you have special comments.

Attachment

Write the combination of nucleic acid donor, donor nucleic acid, vector, host, and possessing organisms along with the containment measures taken in the relevant step of the experiment so that the flow of the experiment is clear. When changing experimental materials, underline the materials to be added.

Table of genetically modified organisms and containment measures.

Nucleic acid donor	Donor nucleic acid	Vector	Host etc.	Possessing organisms	Containment Measure Classification	Note
Streptomyces lavendulae ## strain Mouse E. coli	XX enzyme gene (cDNA) XX enzyme gene (cDNA) lac promoter (genome DNA) Ampicillin-resistant gene	pUC119	JM109 derived from E. coli K12 strain	None	P1	Level B1 Cloning
Mouse Bacteriophage T7 E. coli	XX enzyme gene (cDNA) T7 promoter (genome DNA) Ampicillin-resistant gene lac I (genome DNA)	pET	BL21 (DE3) derived from E. coli B strain	None	P1 Indicate the step of operation.	Level B1 Protein expression
E. coli Schistosoma japonicum Streptomyces lavendulae ## strain	tac promoter (genome DNA) lac I (genome DNA) Glutathione S-transferase(cDNA) XX enzyme gene (cDNA)	pGEX	BL21 derived from E. coli B strain JM109 derived from E. coli K12 strain	None	P1	Level B1 Protein expression