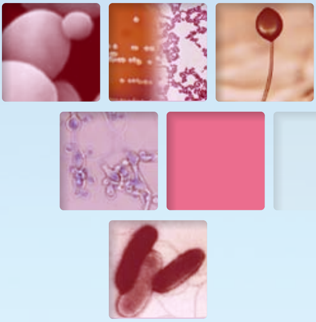
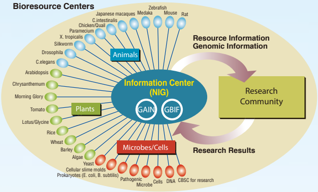
National BioResource Project
2016

Japan Agency for Medical Research and Development

Department of Research Infrastructure



NBRP Information Center Upgrading Program Implementation System



Resource Research Circulation

Introduction

Bio-resources (strains, populations, tissues, cells, genes of animals, plants and microorganisms as research materials) are important infrastructures for life sciences. It is vital that researchers share various bio-resources necessary for pursuing research and development. This is because these resources, produced from years of painstaking labor, form the foundation for future research. Moreover, it is necessary for scientific communities to use a common set of bio-resources so that their research results can be effectively compared. Thus, the development of outstanding collections of bio-resources is essential to give this country an internationally competitive edge in life sciences.

Based on the Science and Technology Basic Plans of the Japanese Government, the Ministry of Education, Culture, Sports, Science and Technology (MEXT) implemented the National BioResource Project (NBRP) in FY2002 to construct the framework for systematic collection, preservation, and distribution of bio-resources, with a focus on those that required strategic development by the national government. Through the revision every 5 years, the NBRP is now in the final year of its third phase and consists of the core centers for 29 categories of bio-resources and a center for information on the resources. Furthermore, the bio-resource framework has been enhanced by increasing value-added genomic resources and developing preservation technologies. Several bio-resource centers have been already recognized to meet the highest global standards.

Based on the Plan for Promotion of Medical Research and Development of the Healthcare Policy approved by the Cabinet in 2014, the operation of the NBRP has been transferred to the Japan Agency for Medical Research and Development (AMED) from FY2015. Currently the Program Supervisor (PS) and the Program Officer (PO) in consultation with the Promotion Committee are responsible for the promotion of the activities of the NBRP, as well as the implementation of its 4th phase, taking into consideration the current trends in life sciences. Finally, we would like to emphasize that the bio-resources in the NBRP cannot be restored once they are lost, which the Great East Japan Earthquake taught us. Your cooperation and support for this project would be highly appreciated.

April 2016

Yuji Kohara, Ph.D.
Program Supervisor, NBRP
(Project Professor, National Institute of Genetics)

List of NBRP Implementing Organizations

Core Facility Upgrading Program

Organism, etc	＊	Principal Investigator	Implementing Organization	Page
Mice	○	Atsushi Yoshiki	Experimental Animal Division, RIKEN BioResource Center	1
Rats	○	Takashi Kuramoto	Institute of Laboratory Animals, Graduate School of Medicine, Kyoto University	2
	B	Atsushi Yoshiki	Experimental Animal Division, RIKEN BioResource Center	
Drosophila	○	Ryu Ueda	Genetic Resource Center, National Institute of Genetics, Research Organization of Information and Systems	3
		Toshiyuki Takano	Drosophila Genetic Resource Center, Kyoto Institute of Technology	
		Masayoshi Watada	Graduate School of Science and Engineering, Ehime University	
		Takeshi Awasaki	Kyorin University, School of Medicine	
	B	Ryo Akashi	Faculty of Agriculture, University of Miyazaki	
C. elegans	○	Shohei Mitani	Tokyo Women's Medical University, School of Medicine	4
Silkworms	○	Yutaka Banno	Institute of Genetic Resources, Faculty of Agriculture, Kyusyu University	5
		Toru Shimada	Graduate School of Agricultural and Life Sciences, The University of Tokyo	
		Zenta Kajiura	Faculty of Textile Science and Technology, Shinsyu University	
		Hideki Sezutsu	Genetically Modified Organism Research Center, National Institute of Agrobiological Sciences	
Medaka	○	Kiyoshi Naruse	National Institute for Basic Biology, National Institutes of Natural Sciences	6
		Mitsuru Sakaizumi	Department of Environmental Science, Niigata University	
	B	Hitoshi Okamoto	RIKEN Brain Science Institute	
	B	Ryo Akashi	Faculty of Agriculture, University of Miyazaki	
Zebrafish	○	Hitoshi Okamoto	RIKEN Brain Science Institute	7
		Koichi Kawakami	Genetic Resource Center, National Institute of Genetics, Research Organization of Information and Systems	
		Shinichi Higashijima	Okazaki Institute for Integrative Bioscience, National Institutes of Natural Sciences	
	B	Kiyoshi Naruse	National Institute for Basic Biology, National Institutes of Natural Sciences	
Japanese macaques	○	Atsushi Nambu	National Institute for Physiological Sciences, National Institutes of Natural Sciences	8
		Katsuki Nakamura	Primate Research Institute, Kyoto University	
Ciona intestinalis	○	Kazuo Inaba	Shimoda Marine Research Center, University of Tsukuba	9
		Yutaka Satou	Graduate School of Science, Kyoto University	
		Koji Akasaka	Misaki Marine Biological Station, Graduate School of Science, The University of Tokyo	
Chicken/Quail	○	Yoichi Matsuda	Avian Bioscience Research Center, Nagoya University	10
Paramecium	○	Masahiro Fujishima	Graduate School of Science and Technology for Innovation, Yamaguchi University	11
Xenopus tropicalis	○	Akihiko Kashiwagi	Institute for Amphibian Biology, Graduate School of Science, Hiroshima University	12
Arabidopsis/Cultured plant cells, genes	○	Masatomo Kobayashi	Experimental Plant Division, RIKEN BioResource Center	13
Rice	○	Yutaka Sato	Genetic Resource Center, National Institute of Genetics, Research Organization of Information and Systems	14
		Toshihiro Kumamaru	Institute of Genetic Resource, Faculty of Agriculture, Kyusyu University	
		Kazuyuki Doi	Graduate School of Bioagricultural Sciences, Nagoya University	
Wheat	○	Shuhei Nasuda	Graduate School of Agriculture, Kyoto University	15
		Kanako Kawaura	Kihara Institute for Biological Research, Yokohama City University	
Barley	○	Kazuhiro Sato	Institute of Plant Science and Resources, Okayama University	16
Algae	○	Masanobu Kawachi	National Institute for Environmental Studies (NIES)	17
		Hiroshi Kawai	Kobe University Research Center for Inland Seas	
		Tsuyoshi Nakayama	Faculty of Life and Environmental Sciences, University of Tsukuba	
	B	Kazuhiro Kogame	Faculty of Science, Hokkaido University	

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Core Facility Upgrading Program

Organism, etc	*	Principal Investigator	Implementing Organization	Page
Chrysanthemum	○	Makoto Kusaba	Laboratory of Plant Chromosome and Gene stock, Graduate School of Science, Hiroshima University	18
Morning glory	○	Eiji Nitasaka	Faculty of Sciences, Kyusyu University	19
		Atsushi Hoshino	National Institute for Basic Biology, National Institutes of Natural Sciences	
Lotus/Glycine	○	Ryo Akashi	Faculty of Agriculture, University of Miyazaki	20
		Jun Abe	Graduate School of Agriculture, Hokkaido University	
		Toshio Aoki	College of Bioresource Sciences, Nihon University	
		Toyoaki Anai	Faculty of Agriculture, Saga University	
Tomato	○	Hiroshi Ezura	Gene Research Center, Faculty of Life and Environmental Sciences, University of Tsukuba	21
		Koh Aoki	Graduate School of Life and Environmental Sciences, Osaka Prefecture Unversity	
Cellular slime molds	○	Hidekazu Kuwayama	Faculty of Life and Environmental Sciences, University of Tsukuba	22
		Taro Ueda	Biomedical Research Institute, National Institute of Advanced Industrial Science and Technology	
	B	Masahiro Ueda	RIKEN Quatitative Biology Center (QBiC)	
Pathogenic microorganisms	○	Takashi Yaguchi	Medical Mycology Research Center, Chiba University	23
		Takayuki Ezaki	Graduate School of Medicine, Gifu University	
		Tetsuya Iida	Research Institute for Microbial Diseases, Osaka University	
		Kenji Hirayama	Institute of Tropical Medicine (NEKKEN), Nagasaki University	
General microbes	○	Moriya Ohkuma	Microbe Division/Japan Collection of Microorganisms (JCM), RIKEN BioResource Center	24
Prokaryotes (E.coli, B.subtilis)	○	Hironori Niki	Genetic Resource Center, National Institute of Genetics, Research Organization of Information and Systems	25
	B	Tsutomu Katayama	Faculty of Pharmaceutical Sciences, Kyusyu University	
Yeast	○	Taro Nakamura	Graduate School of Science, Osaka City University	26
		Minetaka Sugiyama	Graduate School of Engineering, Osaka University	
	B	Kenji Kitamura	Natural Science Center for Basic Research and Development ,Hiroshima University	
DNA material	○	Takehide Murata	Gene Engineering Division, RIKEN BioResource Center	27
Human and animal cells	○	Yukio Nakamura	Cell Engineering Division,RIKEN BioResource Center	28
Cord blood stem cells for research	○	Tokiko Nagamura-Inoue	Department of Cell Processing and Transfusion, Research Hospital, The Institute of Medical Science, The University of Tokyo	29
		Yukio Nakamura	Cell Engineering Division,RIKEN BioResource Center	

* ○ : Core Facility None : Sub Core Facility B : Sub Core Facility for the backup of bioresource

Information Center Upgrading Program

Organism, etc	Core Facility	Principal Investigator	Implementing Organization	Page
Information	○	Yukiko Yamazaki	Genetic Resource Center, National Institute of Genetics, Research Organization of Information and Systems	30
		Tetsuro Matsuzawa	Primate Research Institute, Kyoto University	
		Motomi Ito	Graduate School of Arts and Sciences, The University of Tokyo	
		Tsuyoshi Hosoya	National Museum of Nature and Science	

List of NBRP Implementing Organizations

Genome Information Upgrading Program

Organism, etc	Principal Investigator	Organization	Focus	Project Period	Page
Mice	Toyoyuki Takada	Genetic Strains Research Center, National Institute of Genetics	Genome resequencing of Japanese wild mouse-derived MSM/Ms strain	FY2016	31
Rats	Mikita Suyama	Medical Institute of Bioregulation, Kyushu University	Whole genome resequencing of the representative rat strains and development of a SNP typing kit	FY2016	31
Silkworms	Toru Shimada	The University of Tokyo	Genome Re-sequencing of Diverse Strains of *Bombyx mori* and *B. mandarina* (2)	FY2016	32
Algae	Yuu Hirose	Toyohashi University of Technology	Genome sequencing project of heterocystous cyanobacteria in the NIES collection	FY2016	32
Mice	Yoichi Gondo	RIKEN BioResource Center	Sequence and structure determination and open to public of reference mouse genome based on long one-molecule sequencing.	FY2015	—
Rats	Mikita Suyama	Medical Institute of Bioregulation, Kyushu University	Targeted genome resequencing of 20 strains of the rats	FY2015	—
Drosophila	Shu Kondo	Genetic Resource Center, National Institute of Genetics	Genome sequencing of diverse *Drosophila* species (II)	FY2015	—
Silkworms	Toru Shimada	The University of Tokyo	Genome Re-sequencing of Diverse Strains of *Bombyx mori* and *B. mandarina*	FY2015	—
Lotus	Shusei Sato	Graduate School of Life Sciences, Tohoku University	Generation of high quality genome sequence of Gifu accession of *Lotus japoncius* to accelerate NBRP resource application	FY2015	—
Pathogenic microorganisms	Takashi Yaguchi	Chiba University	Maintenance of whole genome sequences on related species of *Aspergillus fumigatus*	FY2015	—
Rice	Nori Kurata	Genetic Resource Center, National Institute of Genetics, Research Organization of Information and Systems	Generation of genome sequence diversity information for wild relatives of rice	FY2014	—
General microbes	Moriya Ohkuma	Microbe Division/Japan Collection of Microorganisms (JCM), RIKEN BioResource Center	Genome sequencing of eukaryotic microorganisms of NBRP general microbes	FY2014	—
Lotus	Shusei Sato	Graduate School of Life Sciences, Tohoku University	Resequencing of the NBRP collected resources intended to upgrade the genome information of *Lotus japonicus*	FY2014	—
Drosophila	Shu Kondo	Genetic Resource Center, National Institute of Genetics, Research Organization of Information and Systems	Genome sequencing of diverse Drosophila species	FY2014	—
General microbes	Moriya Ohkuma	Microbe Division/Japan Collection of Microorganisms (JCM), RIKEN BioResource Center	Genome sequencing of microbial strains for environmental and health science	FY2012	—
Pathogenic microorganisms	Takayuki Ezaki	GTC Genetic Resource Stock Center of Microbial Pathogens Graduate School of Medicine, Gifu University	Genome Sequencing of Opportunistic Pathogens	FY2012	—
Rat	Tadao Serikawa	Institute of Laboratory Animals, Graduate School of Medicine, Kyoto University	Whole genome sequencing of F344 rat	FY2011	—
Ciona intestinalis/ Oxycomanthus japonicus	Kazuo Inaba	Shimoda Marine Research Center, University of Tsukuba	Genome sequencing of the Ciona intestinalis inbred line	FY2011	—
Mice	Atsushi Yoshiki	Experimental Animal Division, RIKEN BioResource Center	Completion of BAC end sequencing of the mouse C57BL/6N substrain	FY2010	—
Tomato	Koh Aoki	Kazusa DNA Research Institute	Micro-Tom genome sequencing	FY2010	—
Japanese macaques	Tadashi Isa	National Institute for Physiological Sciences, National Institutes of Natural Sciences	Japanese macaque genome sequencing	FY2010	—
Medaka	Kiyoshi Naruse	National Institute for Basic Biology, National Institutes of Natural Sciences	Establishment of polymorphism information of medaka inbred strains	FY2010	—
Mice	Atsushi Yoshiki	Experimental Animal Division, RIKEN BioResource Center	BAC end sequencing of the mouse C57BL/6N substrain	FY2009	—
Medaka	Kiyoshi Naruse	National Institute for Basic Biology, National Institutes of Natural Sciences	Full-length cDNA resources of medaka fish	FY2009	—
Wheat	Yasunari Ogihara	Kihara Institute for Biological Research, Yokohama City Universty	Full-length cDNA resources of common wheat	FY2009	—
Tomato	Erika Asamizu	Gene Research Center, Graduate School of Life and Environmental Sciences, University Tsukuba	Micro-Tom BAC end sequencing	FY2009	—

List of NBRP Implementing Organizations

Genome Information Upgrading Program

Organism, etc	Principal Investigator	Organization	Focus	Project Period	Page
Medaka	Kiyoshi Naruse	National Institute for Basic Biology, National Institutes of Natural Sciences	Medaka Fish Full-length cDNA Resources	FY2008	—
Rats	Tadao Serikawa	Institute of Laboratory Animals, Graduate School of Medicine, Kyoto University	Rat LE/Stm BAC end sequencing	FY2008	—
Tomato	Koh Aoki	Kazusa DNA Research Institute	Enhancing tomato resources by sequencing Micro-Tom full-length cDNA	FY2008	—
Medaka	Kiyoshi Naruse	National Institute for Basic Biology, National Institutes of Natural Sciences	Full-length cDNA resources of medaka fish	FY2007	—
Drosophila	Ryu Ueda	Genetic Strains Research Center, National Institute of Genetics, Research Organization of Information and Systems	Genome and property information for the quality control of *Drosophila* strains	FY2007	—
Arabidopsis	Masatomo Kobayashi	Experimental Plant Division, RIKEN BioResource Center	Sequence analysis of full-length cDNAs of *Thellungiella halophila* as new *Arabidopsis* resources	FY2007	—
Wheat	Yasunari Ogihara	Kihara Institute for Biological Research, Yokohama City University	Full-length cDNA resources of bread wheat	FY2007	—

Fundamental Technology Upgrading Program

Organism, etc	Principal Investigator	Organization	Focus	Project Period	Page
Drosophila	Toshiyuki Takano	Kyoto Institute of Technology	Development of a new cryopreservation method for Drosophila stocks	FY2016	33
C. elegans	Shohei Mitani	Tokyo Women's Medical University School of Medicine	Construction of High-Performance balancers for *C. elegans*	FY2016	33
Rats/Zebrafish/ X. tropicalis	Takashi Yamamoto	Graduate School of Science, Hiroshima University	Development of easy protocols for efficient gene knock-in using genome editing technology	FY2016	34
Mice	Atsushi Yoshiki	Experimental Animal Division, RIKEN BioResource Center	Fundamental technology development of genome editing for the establishment of intractable disease models	FY2016	34
Silkworms	Yutaka Banno	Institute of Genetic Resources, Faculty of Agriculture, Kyusyu University	Development of cryopreservation methods of the silkworm	FY2014	—
Mice	Fumihiro Sugiyama	Laboratory Animal Resource Center, University of Tsukuba	Development of Cre-loxP recombination atlas for Cre-driver mouse strains	FY2014	—
Mice	Naomi Nakagata	Center for Animal Resources & Development (CARD), Kumamoto University	Establishment of *in vitro* fertilization systems for all mouse strains	FY2012-2013	—
Medaka	Goro Yoshizaki	Graduate School of Marine Science and Technology, Tokyo University of Marine Science and Technology	Production of functional gametes derived from cryopreserved germ-line stem cells using a surrogate broodstock system in medaka	FY2012-2013	—
Drosophila	Ryu Ueda	Genetic Resource Center, National Institute of Genetics, Research Organization of Information and Systems	Development of cryopreservation method of Drosophila strains	FY2012-2013	—
Rats	Tadao Serikawa	Institute of Laboratory Animals, Graduate School of Medicine, Kyoto University	Improving the efficiency of sperm preservation technologies in rats	FY2010-2011	—
Mice	Yasumasa Ishida	Graduate School of Biological Sciences, Nara Institute of Science and Technology	Production of conditionally gene-disrupted ES-cell clones and establishment of a database for the inactivated genes	FY2010-2011	—
Drosophila	Masa-Toshi Yamamoto	Drosophila Genetic Resource Center, Kyoto Institute of Technology	Development of a long-term stable preservation technology for Drosophila strains	FY2007-2009	—
Medaka	Minoru Tanaka	National Institute for Basic Biology, National Institutes of Natural Sciences	Development of standard strains for the functional analysis of medaka genes	FY2007-2009	—
DNA (animal/plant/ microbial)	Masatomo Kobayashi	Experimental Plant Division, RIKEN BioResource Center	Development of long-term preservation technology for genetic resources	FY2007-2009	—
Mice	Yasumasa Ishida	Graduate School of Biological Sciences, Nara Institute of Science and Technology	A novel gene-disruption strategy based on the suppression of NMD	FY2007-2008	—
Mice/Rats	Atsushi Yoshiki	Experimental Animal Division, RIKEN BioResource Center	Development of transportation systems for mouse and rat resources	FY2007-2008	—

National BioResource Project

National BioResource Project

Purpose

The major purpose of the National BioResource Project (NBRP) is to collect, preserve, and provide bioresources (such as experimental animals and plants) that are essential experimental materials for life science research. In order to meet current scientific demands, the project also aims to increase the value of bioresources via addition of genome information and development of fundamental technologies for preservation and other necessary procedures. In addition, the information center will be upgraded in order to promote dissemination of information regarding the whereabouts and biological characteristics of bioresources.

Background

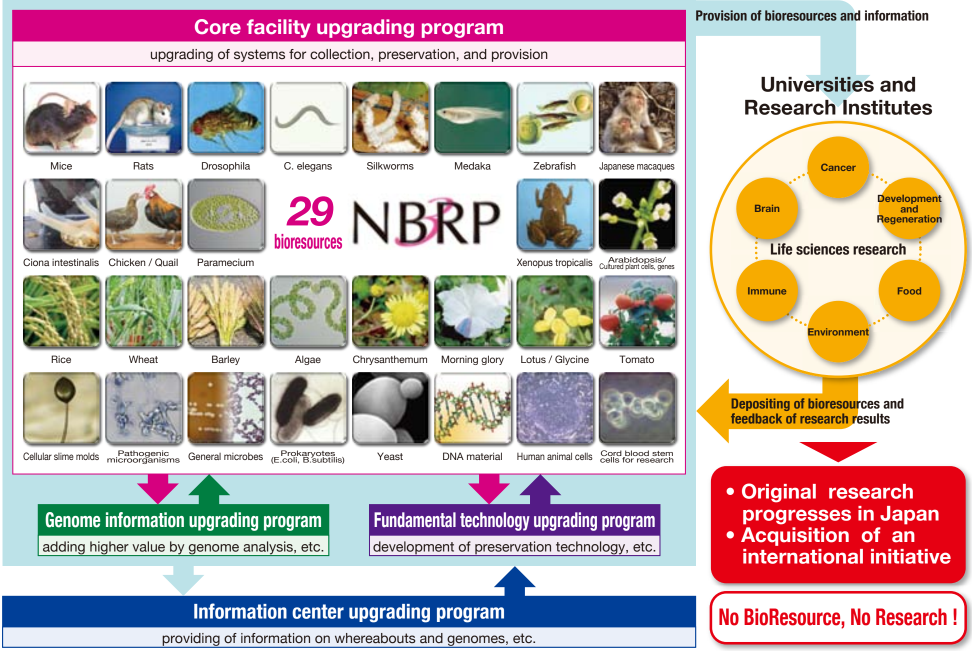
Based on the Plan for Promotion of Medical Research and Development of the Healthcare Policy approved by the Cabinet in 2014, operation of the NBRP was transferred to the Japan Agency for Medical Research and Development (AMED) from the Ministry of Education, Culture, Sports, Science and Technology (MEXT) in FY2015.

In the NBRP from FY2002 to FY2016, systems for collection, preservation, and provision were established for bioresources such as experimental animals, plants, and microbes that are important to promote life science research.

In the Fifth Science and Technology Basic Plan approved by the Cabinet in 2016, the government stipulated that intellectual infrastructures such as biological and genetic resources should be collected and utilized by public organizations both strategically and systematically. Therefore, the NBRP plans to reinforce existing intellectual infrastructures, by improving the quality of the available resources in response to diverse needs.

Under these circumstances, AMED continues to operate the NBRP to promote strategic collection and utilization of the bioresources.

Project Aims



Core facility upgrading program

upgrading of systems for collection, preservation, and provision

Mice, Rats, Drosophila, C. elegans, Silkworms, Medaka, Zebrafish, Japanese macaques

Ciona intestinalis, Chicken / Quail, Paramecium, 29 bioresources NBRP, Xenopus tropicalis, Arabidopsis/ Cultured plant cells, genes

Rice, Wheat, Barley, Algae, Chrysanthemum, Morning glory, Lotus / Glycine, Tomato

Cellular slime molds, Pathogenic microorganisms, General microbes, Prokaryotes (E.coli, B.subtilis), Yeast, DNA material, Human animal cells, Cord blood stem cells for research

Genome information upgrading program

adding higher value by genome analysis, etc.

Fundamental technology upgrading program

development of preservation technology, etc.

Information center upgrading program

providing of information on whereabouts and genomes, etc.

Provision of bioresources and information

Universities and Research Institutes

Life sciences research

Cancer, Development and Regeneration, Food, Environment, Immune, Brain

Depositing of bioresources and feedback of research results

- Original research progresses in Japan
- Acquisition of an international initiative

No BioResource, No Research !

Project General Outline

The National BioResource Project implements the following four programs to facilitate the collection, preservation, and provision of bioresources and the development of related technologies; (1) core facility upgrading program, (2) genome information upgrading program, (3) fundamental technology upgrading program, and (4) information center upgrading program. These programs will accomplish the purpose of the project described above and will be coordinated to produce synergistic effects.

(1) Core facility upgrading program:

The core facilities will be established to carry out collection, preservation, and provision of bioresources. The bioresources selected for the NBRP are of fundamental importance in life sciences research and must be excellent and originally produced in Japan.

(2) Genome information upgrading program:

The aim of this program is to improve quality and increase the value of bioresources, as well as to reinforce the uniqueness and leading position of Japanese bioresources by enriching and expanding strain and characteristics information, genetic information, such as genome sequences of cDNA, and genome resources, including genome libraries of bioresources that are collected, preserved, and provided by the NBRP.

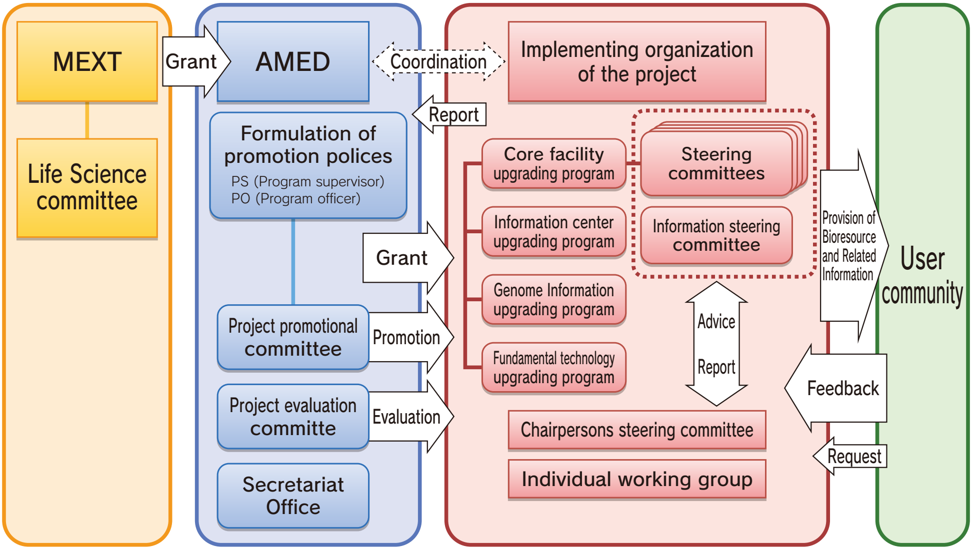
(3) Fundamental technology upgrading program:

Development of technologies relating to collection, proliferation, quality management, preservation and provision of bioresources which are the subjects of the core facilities promotion program is conducted.

(4) Information center upgrading program:

Construction of databases of whereabouts information, genetic information, and biological characteristics of bioresourcesthat are gathered at the core facilities, and public relations of the NBRP through its home page is upgraded.

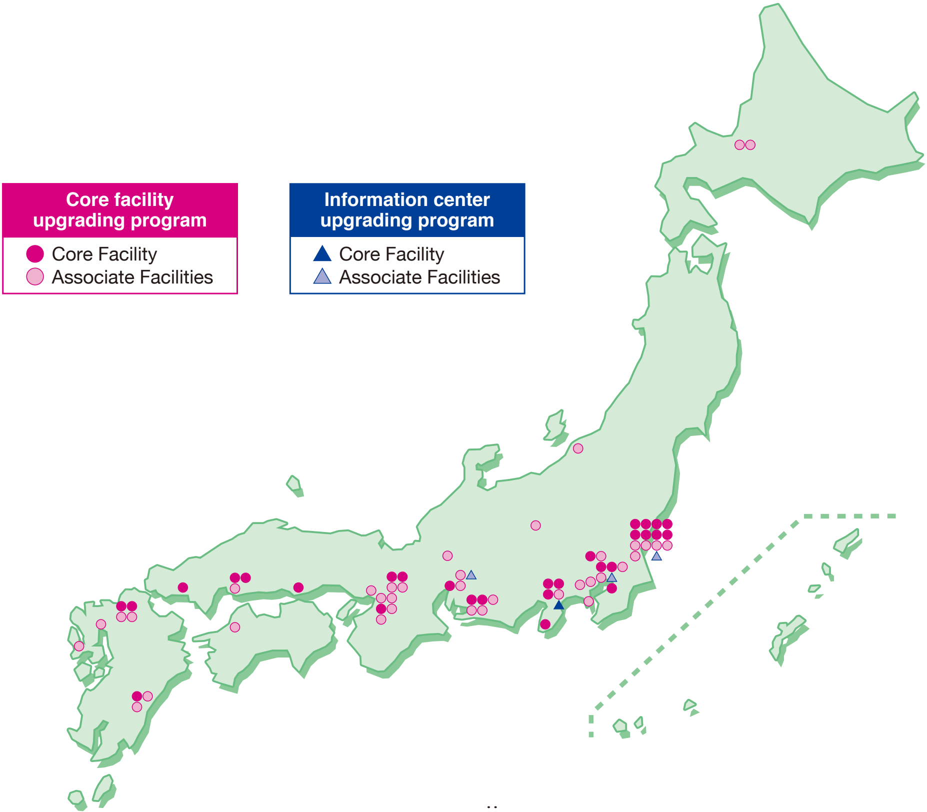
Project Implementation System



NBRP Promotion Committee

Position	Name	Affiliated organization
Chairman	Yuji Kohara	Professor, National Institute of Genetics (NIG)
Vice-Chairman	Yuichi Obata	Director, RIKEN BioResource Center (BRC)
	Hideko Urushihara	Professor Emeritus, University of Tukuba
	Kiyotaka Okada	Professor, Faculty of Agriculture, Ryukoku University
	Motoya Katsuki	Deputy Director, Research Center for Science Systems, Japan Society for the Promotion of Science (JSPS)
	Makoto Kawase	Professor, Facuty of Life and Environmental Sciences, University of Tukuba
	Kazuo Shinozaki	Director, RIKEN Center for Sustainable Resource Science (CSRS)
	Toshihiko Shiroishi	Vice-Director · Professor, National Institute of Genetics (NIG)
	Tetsuya Hayashi	Professor, Graduate School of Medical Sciences, Kyushu University
	Hiroo Fukuda	Director/Professor, Graduate School of Sciences, The University of Tokyo

NBRP Network of Japan



CORE FACILITY UPGRADING PROGRAM Mice

Core Facility : Experimental Animal Division, RIKEN BioResource Center
Principal Investigator : Atsushi Yoshiki FAX : +81-29-836-9010
Contact site : animal@brc.riken.jp
URL : http://mus.brc.riken.jp/en/

Overview

Mice are essential model animals to understand our health and conquer complex diseases. RIKEN BioResource Center (BRC) has operated as an international hub of mouse resources to collect, preserve and distribute mouse models created in Japan for the study of gene functions and human diseases. To meet research community's needs, our mice are cleaned-up to specific pathogen-free state, strictly monitored for their health, and accurately tested on their genetic modifications and backgrounds. Genomic, gene expression and phenotypic information are added to enrich their value to establish mouse resources of the world highest standard. We promote backup-storage of frozen embryos and sperm to protect our resources against disasters.

RIKEN BRC participates in the International Mouse Strain Resource, IMSR and registers strains created by Japanese scientists and disseminate the mice around the world. We also promote Asian/Australian networks to strength regional cooperation. Moreover, we have collaborated with the Japan Mouse Clinic and participated in the International Mouse Phenotyping Consortium (IMPC) to contribute to basic medical sciences and drug discovery by producing knockout mice for every coding gene, generating broad-based phenotypic data, and making them available to scientists around the world.

Key Strains/Studies

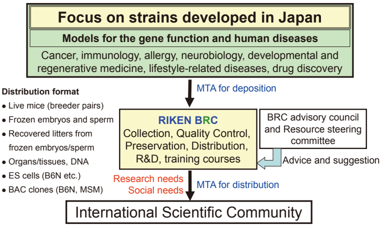
- **C57BL/6-*App*$^{tm1(NL)Tcs}$/TcsRbrc (RBRC06342)**
- **C57BL/6-*App*$^{tm2(NL-F)Tcs}$/TcsRbrc (RBRC06343)**
- **C57BL/6-*App*$^{tm3(NL-G-F)Tcs}$/TcsRbrc (RBRC06344)**

Drs. Saido, Saito, and colleagues at RIKEN Brain Science Institute have developed the next generation mouse models for Alzheimer's disease (AD) by knock-in to the App gene with Swedish (NL), Iberian (F) and Arctic (G) mutations found in familial AD patients. This mouse model well recapitulates patients' amyloid pathology (Fig.1), being used in over 150 laboratories around the world and is expected to become a standard model to find preventive therapies of the AD.

- **C57BL/6N-*Gt(ROSA)26Sor*$^{tm1(CAG-EGFP/DsRed)Utr}$/ (R26GRR, RBRC04874)**
- **C57BL/6J-*Ins1*$^{em1(cre)Utr}$ (RBRC09525)**

To improve the quality of Cre-driver mice, "Development of Cre-loxP recombination atlas for Cre-driver mouse strains" was conducted in the NBRP Fundamental Technologies Upgrading Program in FY2014 by Dr. Sugiyama, University of Tsukuba to characterize Cre-driver strains using R26GRR reporter mice (RBRC04874) which enable evaluating the tissue specificity of Cre-recombinase expression. These Cre-expression data will be added to upgrade the quality of Cre-driver strains.

Operational structure for mouse resources



Focus on strains developed in Japan

Models for the gene function and human diseases

Cancer, immunology, allergy, neurobiology, developmental and regenerative medicine, lifestyle-related diseases, drug discovery

MTA for deposition

Distribution format

- Live mice (breeder pairs)
- Frozen embryos and sperm
- Recovered litters from frozen embryos/sperm
- Organs/tissues, DNA
- ES cells (B6N etc.)
- BAC clones (B6N, MSM)

RIKEN BRC
Collection, Quality Control, Preservation, Distribution, R&D, training courses

BRC advisory council and Resource steering committee

Advice and suggestion

Research needs
Social needs

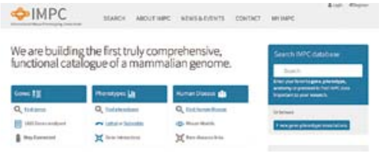
MTA for distribution

International Scientific Community

International collaboration

International Mouse Strain Resource,
IMSR One-Stop Shop Database
http://www.findmice.org/

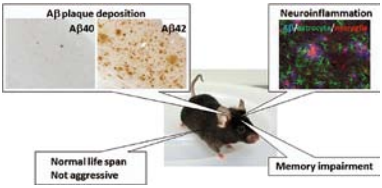
Asian Mouse Mutagenesis
Resource Association, AMMRA
http://www.ammra.info/



IMPC

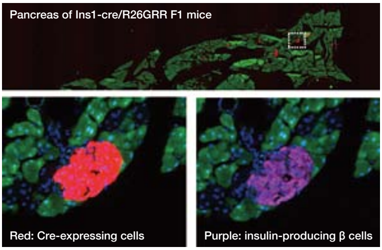
We are building the first truly comprehensive, functional catalogue of a mammalian genome.

International Mouse Phenotyping Consortium, IMPC
http://www.mousephenotype.org/



Courtesy of Takaomi C. Saido, Ph.D. and Takashi Saito, Ph.D.

Fig. 1. Alzheimer's disease model with human patients' mutations
Nat Neurosci 17, 661-3, 2014



Courtesy of Fumihiro Sugiyama, Ph.D.

Fig. 2. Pancreatic β cell-specific Cre recombinase expression in the Ins1-cre mice

Fig. 2. Upper: A fluorescent image of the pancreatic tissue of Ins1-cre/R26GRR F1 mice by crossing Ins1-cre (RBRC09525) x R26GRR reporter (RBRC04874) mice. Lower left: High magnification image of the above. In Cre-recombinase expressing cells, GFP (green) was excised to express DsRed (red). Non-recombinant cells remained to express GFP (green). Cell nuclei were labelled in blue with DAPI. Lower right: pancreatic β cells were labelled with anti-human insulin antibody in purple.

CORE FACILITY UPGRADING PROGRAM Rats

Core Facility : Institute of Laboratory Animals, Graduate School of Medicine,
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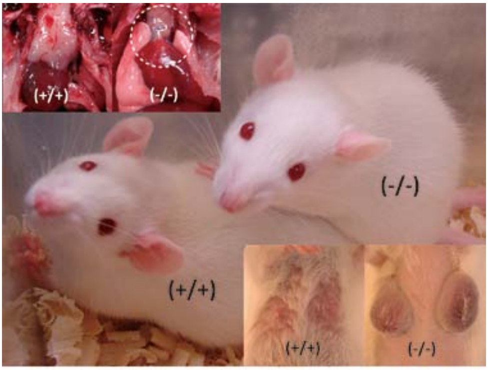
Overview

The rat is the mammal which is used in many fields of research owing to its suitable size, adaptability and neurological characteristics. Recent developments including the establishment of rat ES/iPS cells and the generation of gene knockout rats using gene editing nucleases (ZFN/TALEN/CRISPR) technology etc. will boost the utility of the rat as biological resource.

The Institute of Laboratory Animals, Graduate School of Medicine, Kyoto University, conducts a rat strain-based collection, preservation and distribution program and quality assurance through microbial and genetic monitoring, enhances rat strain databases and holds the Rat Resource Research Meeting to support and stimulate science in the rat research community. Riken BRC, backs up frozen embryos and sperm to support the central facility at Kyoto University as a sub center.

The NBRP-Rat has been developed with the intent of being the world's leading rat resource center. This project promotes further utilization of the rat as a research tool in many fields of science.

Various rat strains deposited to NBRP-Rat



X-SCID Rat (+/+ : Wild type、-/- : *Il2rg* mutated)
Left : Lack of thymus、Right : Xenoplantation of human tumor cells

Key Strains/Studies

So far, 802 different strains have been deposited to NBRP-Rat and animals have been supplied to researchers in 1,104 cases. The repository includes spontaneous mutants, recombinant inbred, congenic, consomic, transgenic, and knock out rats. These strains are utilized in fields as neurobiology, cardiovascular disease/hypertension, diabetes/obesity, cancer, immunology, development and metabolism.

● Severe combined immunodeficiency rats (X-SCID, SCID, FSG)

Immunodeficient rats were established using gene editing nucleases (ZFN/TALEN). These strains can act as hosts for human xenogeneic tissue grafts and stem cell transplantation.

● Reporter gene transgenic rats

GFP, DsRed, LacZ and other marker genes are important tools for the examination of many biological processes. Our repository has many of such marker strains available for various kinds of experiments with ubiquitous or organ specific marker expression.

● KURMA (Kyoto University Rat Mutant Archive)

Sperm and DNA of 10,752 ENU mutagenized F344 G1 animals are integrated into the NBRP-Rat. This mutant archive, KURMA10K, provides gene-targeted rats as animal models for various fields in biomedical research.

CORE FACILITY UPGRADING PROGRAM Drosophila

Core Facility : Genetic Resource Center, National Institute of Genetics
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Overview

The purposes of this program are to comprehensively maintain, manage, and widely distribute to research communities the genetic resources of *Drosophila*, such as (1) mutant strains of *Drosophila melanogaster*, which are useful as a basis or platform for life science studies, and (2) mutant strains of the wild species of *Drosophila* or related species of *Drosophila melanogaster*, which are important for evolution and biodiversity studies. To this end, five organizations —the National Institute of Genetics, Kyoto Institute of Technology, Ehime University, Kyorin University, and Miyazaki University— are to constitute a consortium for the joint project. By the second stage in the next 10 years, the consortium aims to assume international responsibility as the fully developed, world's largest stock center by collecting the resources and improving the quality according to the needs of the times; thus we will contribute to the acceleration of leading-edge research activities in user communities.

Key Strains/Studies

National Institute of Genetics

- RNAi mutant strains (15,522 for 7,800 genes)

Kyoto Institute of Technology

- Basic strains (3,500) ● NP strains (4,200)
- GS strains (7,000) ● FRT-lethal strains (1,300)
- MARCM strains (2,200) ● DrosDel strains (520)
- Imaging strains (1,700) ● RS strains (2,800)
- Others (3,000)

Ehime University

- Japanese wild-type strains (54 species, 694 strains)
- Asian wild-type strains (40 species, 156 strains)
- Others (59 species, 254 strains)

Kyorin University

- Mutant strains of species other than *D. melanogaster* (845)
- anapal interspecific mosaic genome lines (266)
- sequenced species (12 species 17strains)

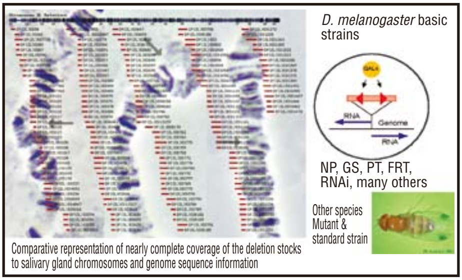
Proteomics Research

In *Drosophila*, by using chromosomal rearrangements, a small Drosophila population of genetically identical background can be constructed in the laboratory. The genetically well manipulated and homogenous flies, which were reared by the breeding techniques, can be applied widely to the proteomics research on various organs, tissue cells and lymphocytes. (Nature Genetics 38: pp. 1440–1445, 2006; Proteomics 9: pp. 2484–93, 2009; doi: 10. 1016/j. jprot. 2010. 05. 006, 2010)

Stock maintenance and strains

Drosophila melanogaster

Drosophila culture: culture bottle & fly stocks in incubator



Comparative representation of nearly complete coverage of the deletion stocks to salivary gland chromosomes and genome sequence information

Some examples among extensive database

Speciation Study

By applying the advanced *Drosophila* research techniques to the classical evolutionary studies such as lethality, sterility and sex ratio distortion resulted in terms of interspecific hybrids between closely-related species, understanding of the causative genes and the speciation mechanisms can be anticipated. (Science 326: pp. 1538–41, 2009; Science 323: pp. 779–82, 2009: Science 323: pp. 376–9, 2009)

CORE FACILITY UPGRADING PROGRAM *C. elegans*

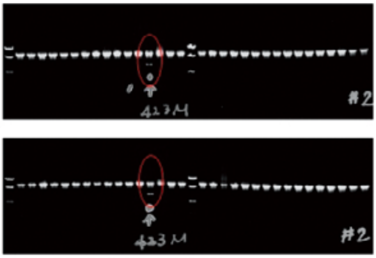
Core Facility : Tokyo Women's Medical University School of Medicine
Principal Investigator : Shohei Mitani FAX : +81-3-5269-7414
Contact site : mitani.shohei@twmu.ac.jp
URL : http://shigen.lab.nig.ac.jp/c.elegans/

Overview

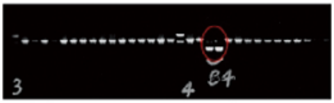
One of the objectives of Phase 3 of the Ministry of Education, Culture, Sports, Science and Technology's NBRP, which began in FY2012, is to advance the collection, preservation, and distribution of *Caenorhabditis elegans* mutants. *C. elegans* is a good model organism for life science research. Genome and EST information has elucidated the gene composition of nearly all chromosomes, and that gene data will be used to collect deletion mutants through reverse genetics. This project is aimed at supporting genetic research on *C. elegans* by collecting and organizing numerous deletion mutants.

The core facility for this project, Tokyo Women's Medical University School of Medicine, already has a repository of cryopreserved strains with random mutations covering nearly all genes. A tube containing the target mutant is identified through PCR (Fig. 1), and the mutant is checked for the deletion before it is published and supplied to users (Fig. 2).

Fig. 1

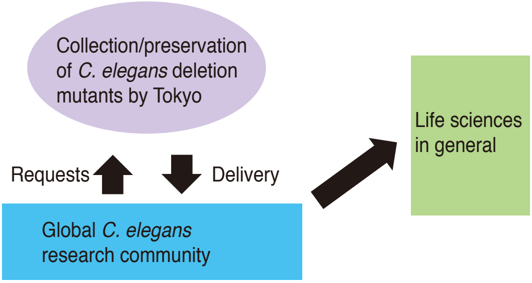


(1) Primary screening



(2) Secondary screening

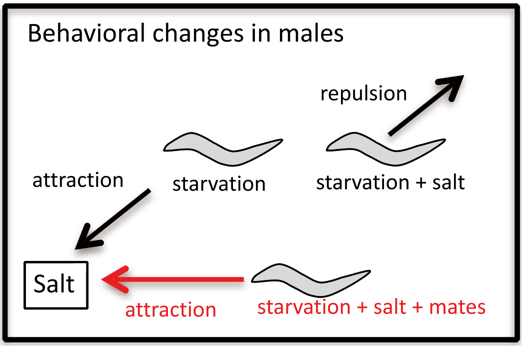
Fig. 2 : System of research using *C. elegans* deletion mutants



Key Strains/Studies

● *pdf-1 (tm1996)*

One of the leading fields of research using *C. elegans* is analysis of learning and memory. Sammut *et al* (Nature **526**, 385 (2015)) used a *pdf-1* (*tm1996*) mutant and showed that the gene is required for male sex-specific learning that animals are attracted to salt even if conditioned with salt and without food (Figure). The authors demonstrated that this learning is regulated by neurons MCM which are derived from glia by transdifferentiating to neurons at the larval stage.



CORE FACILITY UPGRADING PROGRAM Silkworms

Core Facility : Graduate School of Agriculture, Kyushu University
Principal Investigator : Yutaka Banno TEL/FAX : +81-92-624-1011
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Overview

Silkworms are unable to live in the wild and can only survive under human care. In other words, the silkworm is a highly conditioned organism, which is ideal for comparative research between it and its relatives, including the wild silkmoth species thought to be its progenitor. Such comparative investigation should shed light on the mechanisms of conditioning as well as bioutilization and bioevolution. As it is well known, the silkworm played an important agricultural role in supporting Japan’s modernization, and traditionally, Japan has led the world as a repository of silkworm bioresources. Phase 1and 2 of the NBRP focused on upgrading the quality of these resources and consolidating them in a core facility as well as establishing a system for facilitating research. When Phase 1 was initiated, there were inadequacies in the availability of genome information and transgenic technology, but the ability to draw on the outcome of research in other projects has improved the research environment, increasing the need to upgrade and strengthen the resources, which serve as the starting point for research. In Phase 3 the organizations listed below will handle specific projects aimed at establishing the world’s highest-quality resources and making them available globally. The Information Center Upgrading Program will help provide a more solid research base by enabling us to disseminate genetic information about silkworm bioresources and to offer resource information to academic and research associations.

Roles of organizations participating in Phase 3

○ **Collection, maintenance and supply of silkworm strains (Kyushu University)**
  - ● Creation of a world-class collection of mutant strains, upgrading and expansion of collection of strains, and improvement in quality of strains maintained.
  - ● Establishment of a method of safe and efficient maintenance of resources through cryopreservation of germplasm and practical application of artificial insemination technology
  - ● Establishment of a system to supply silkworm strains year round

○ **Collection, maintenance and supply of wild strains (Shinshu University)**
  - ● Collection, maintenance and supply of wild strain resources, such as *Antheraea yamamai*, *Antheraea pernyi*, *Samia cynthia pryeri*

○ **Collection, maintenance and supply of genome resources (Tokyo University)**
  - ● Upgrading and efficient maintenance of cDNA clones and BAC library and improving accessibility

○ **Collection and maintenance of genetically modified silkworms (National Institute of Agrobiological Sciences)**
  - ● Collection and evaluation of genetically modified silkworms Wild silkmoth mimicking its surroundings in the wild; photo at right shows varieties

Wild silkmoth mimicking its surroundings in the wild; photo at right shows varieties of mutant silkworm larvae

Key Strains/Studies

● p50 strain used in genome information analysis
● w-strain collection with multiple chromosome identification markers

The silkworm cocoons shown have not been dyed, but are the result of genetic mutation. Recent research on yellow and white cocoons has elucidated one of the causes of cocoon pigmentation. Silkworms that make yellow cocoons have a protein that binds with, and transports, the yellow pigment (carotenoids) contained in mulberry leaves into their bodies, enabling them to absorb carotenoids from mulberry trees and make yellow cocoons. With white cocoons, the gene for the same protein is defective. To clarify this, genetic recombination technology was used to insert the gene from the silkworm that makes yellow cocoons into the silkworm that makes white cocoons, resulting in yellow cocoons. Carotenoids exist in many living organisms, including humans, and they are essential for such structures as the retina in humans, but the mechanism for their transportation has not been well understood. The results of this research should be useful for understanding the other mutant cocoon colors, such as pink and green. This research highlights the extensive commonality found in biological phenomena and the importance of accumulating the results of research that cross the species boundary.

Sakudoh et al., (2007) PNAS, vol. 104, (21), 8941.

CORE FACILITY UPGRADING PROGRAM **Medaka**

Core Facility : National Institute for Basic Biology
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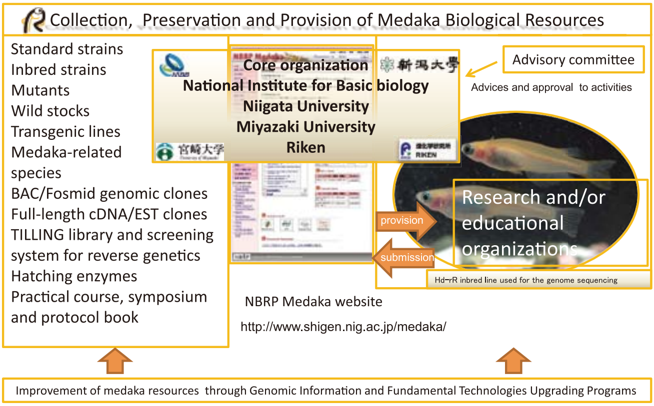
URL : http://www.shigen.nig.ac.jp/medaka/
http://www.nibb.ac.jp/bioresources/

Overview

Medaka has been used as a biological material for more than 100 years in Japan. Our forerunners continuously developed medaka bio-resources and preserved them over this extensive period. Several inbred lines including the Hd-rR line whose sex can be distinguished by body color differences, wild stocks, transgenic lines, mutants with specific phenotypes and medaka-related species are some examples of these resources. Recent achievements in medaka genome analysis provide us with a rich source of genomic information such as full-length cDNA/EST clones and BAC/Fosmid clones covering 90% of the medaka genome. Draft level genome sequence information has also become available. Four organizations, National Institute for Basic Biology (NIBB), Niigata University, Miyazaki University and Riken were selected as the core organization of NBRP medaka by MEXT Japan in 2012 and continue to collect, preserve and provide these medaka bio-resources. This project aims to establish world-class biological resources to promote the utilization of medaka in a wide range of settings from elementary school education to advanced biology and biomedical research. NIBB collects, preserves, and provides the standard stocks, inbred strains, mutants and transgenic lines as well as genomic resources such as full-length cDNA/EST clones and BAC/Fosmid clones and hatching enzyme. We also provide the TILLING library and it's screening system by high resolution melting method and genome editing platform using CRISPR-Cas9 including the construction of gRNA plasmid and microinjection of gRNA with Cas7 mRNA. These services enhance the reverse genetic approach using medaka. Niigata University collects, preserves, and provides wild stocks and medaka-related species and also preserves the inbred strains for backup. Miyazaki University and Riken play an active role in the backup for the genomic resources and the cryopreserved sperm stocks.



Key Strains/Studies

Maintains and supplies the following strains:

- **d-rR** (sex can be distinguished by body color)
- **transparent strains** (Quintet, STII, STIII)
- **Inbred strains** (Hd-rR, HNI, Kaga, HSOK, etc.)
- **Wild stocks** (all 66 lines maintained as live stock)
- **transgenic strains** (d-rR-Tg (beta-actin-loxP-DsRed2-loxP-GFP), expressing DsRed2 in whole body (DsRed2 being converted to GFP with cre expression), Tg (CMV-H2B-GFP), expressing H2B-GFP fusion protein to visualize chromosomes, hse-cre: mcherry heat inducible cre-mcherry fusion protein for the loxP site specific recombination Related species (*O. curvinotus*, *O. dancena*, *O. javanicus* (known as marine medaka), etc.).

Genome resources: 18000 independent full-length cDNA/EST; BAC/Fosmid clones covering 90% of the medaka genome; Hatching enzymes essential for embryonic manipulation. TILLING library and it's screening system by HRM method and genome editing platform using CRISPR-Cas9 for reverse genetic approach.

Identification of sex determining genes, Whole genome sequencing using Hd-rR and HNI strains, Positional cloning of causal genes of mutants, b (orange-red variety), i-3 (albino), pc (cystic kidney disease), ktu (randomization of left-right asymmetry mutant) and application to human disease models such Non-alcoholic fatty liver syndrome model, Parkinson disease and osteoporosis. Identification of ovarian stem cell and a germ cell-intrinsic factor for sperm-egg fate decision in vertebrates. Molecular and neural mechanism of mating preference.

CORE FACILITY UPGRADING PROGRAM Zebrafish

Core Facility : RIKEN Brain Science Institute
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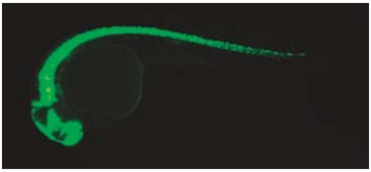
Overview

Zebrafish is widely used as a model vertebrate in which genetic approaches can be performed. The genomic information and techniques for embryology have been accumulated and now zebrafish plays an important role in basic biology and also a model for human diseases.

The number of zebrafish researchers in Japan is increasing. Accordingly, the number of mutant lines and transgenic lines made in Japan is also rapidly increasing. Under these circumstances, the major aim of this project is to set up a system for collecting, maintaining and distributing fish lines for the following purposes: (1) to supply researchers in Japan with lines of their interests quickly. (2) to supply researchers in foreign countries with zebrafish lines created in Japan to increase Japan's contribution to the community.

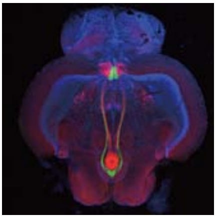


Key Strains/Studies

RIKEN (Hitoshi Okamoto)

Strain: dao:cre-mCherry; vglut2a:loxP-DsRed-loxP-GFP

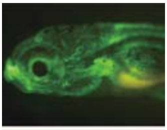
Habebulo-raphe pathway (shown in green), a conserved neural circuit among the vertebrates, encodes the expected level of aversiveness for learning appropriate behavior to avoid the danger.
Amo et al., Neuron, 87 1034-1048 (2014)

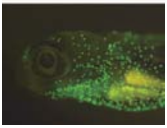


National Institute of Genetics (Koichi Kawakami)

Strain: 600 Gal4-expressing transgenic lines and UAS lines

In these lines, a modified yeast transcription factor Gal4FF is expressed in specific tissues, cells and organs. By crossing these with UAS fish, a desired gene can be expressed in a desired place.
Asakawa, K. et al. Genetic dissection of neural circuits by Tol2 transposon-mediated Gal4 gene and enhancer trapping in zebrafish. Proc. Natl. Acad. Sci. USA 105, 1255-1260 (2008)

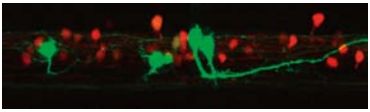




National Institutes of Natural Sciences (Shinichi Higashijima)

Strain: chx10:loxP-DsRed-loxP-GFP

This strain uses the Cre-loxP system. Normally, DsRed is expressed in all alx-positive cells, but by using Cre, it is possible to express EGFP instead of DsRed in some (or all) alx-positive cells. Kimura et al., J Neurosci. May 24; 26:5684-97. 2006.



CORE FACILITY UPGRADING PROGRAM Japanese macaques

Core Facility : National Institute for Physiological Sciences
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URL : http://nihonzaru.jp/

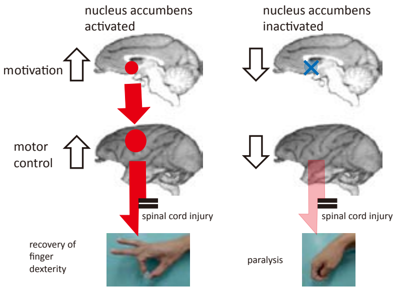
Overview

The Japanese macaque is a middle-sized monkey similar to the Rhesus macaque and the Long-tailed macaque. These are all classified into the genus *Macaca* and belong to the *Cercopithcinae*. Monkeys of the genus *Macaca*, so-called macaques, are relatively close to humans and are indispensable experimental animals for research on higher brain functions, infections/immunology, and regenerative medicine.

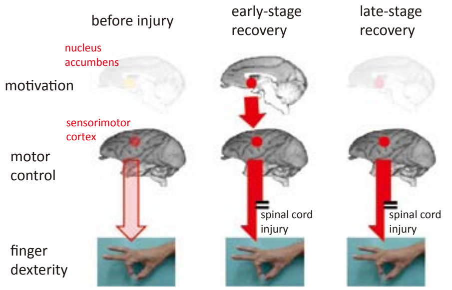
The Japanese macaques, which are indigenous to Japan, have often been used in the fields of neuroscience and physiology in Japan. They have a very high level of curiosity and are temperate in nature. They also have less genetic mutation than other macaques that inhabit a wide area throughout Southeast Asia. Because the amount of ecological, behavioral, genetic and morphological literature available concerning Japanese macaques is the largest for all monkey species, it is regarded as an extremely useful experimental animal.

Until the start of Phase 1 of NBRP, there had been no major purpose-breeding plan for Japanese macaques. However, given future research trends, there is an obvious need for monkeys raised under supervision with records of age, growth process and genealogy. This is the reason why the current project was started as a joint effort between neuroscientists and primate researchers.

In Phase 3, the NBR steering committee, established within the National Institute for Physiological Sciences, keeps promoting the project, jointly with the Kyoto University Primate Research Institute (sub-center).



Inactivation of the neucleus accumbense during the early recovery period diminished high-frequency oscillatory activity in the sensorimotor cortex, which was accompanied by a transient deficit of amelioration in finger dexterity obtained by rehabilitation.



The nucleus accumbens, a motivation center, is directly involved in the control of finger movements during recovery after spinal cord injury.

©2015 NIPS, (Nishimura, et al. Science 2015)

Key Strains/Studies

The following are findings obtained by studies using Japanese macaques:

- the development of pathway-selective and reversible transmission blocking technique by virus vectors (Isa et al. Nature 2012).
- the mechanism of deep brain stimulation (DBS) to alleviate the symptoms of Parkinson's disease (Nambu et al. The Journal of Neuroscience 2013).
- the finding of pulvinar neurons responding strongly and faster to snake images as neuroscientific evidence in support of the Snake Detection Theory (Isbell, 2006) for evolution of the primate brain (Le, Nishijo, et al. PNAS 2013).
- the direct neurophysiological evidence for psychological models of dual-task interference and capacity limitation (Watanabe and Funahashi, Nature Neuroscience 2014).
- the potential contribution of the nucleus accumbens to movement control after spinal cord injury (Sawada et al. Science 2015).
- the first demonstration of modeled vocal tics in Tourette syndrome utilizing PET imaging (McCairn et al. Neuron 2016).

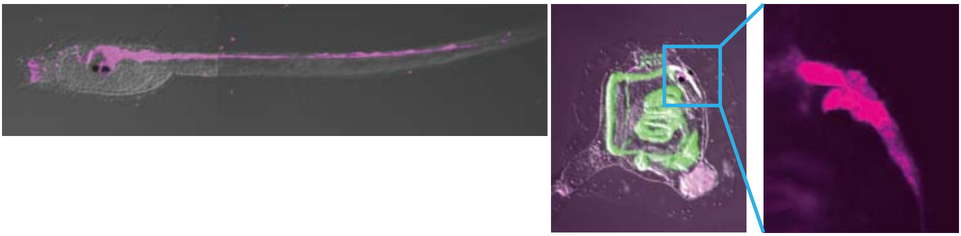
CORE FACILITY UPGRADING PROGRAM Ciona intestinalis

Core Facility : Shimoda Marine Research Center, University of Tsukuba
Principal Investigator : Kazuo Inaba FAX : +81-558-22-0346
Contact site : kinaba@kurofune.shimoda.tsukuba.ac.jp
URL : http://www.shimoda.tsukuba.ac.jp/

Overview

Marine invertebrates have long been regarded as excellent materials for research on the embryogenesis, evolution, reproduction and neurophysiology of animals. Two marine organisms, the urochordate ascidian *Ciona intestinalis*, has been selected as objects of the National BioResource Project.

Ciona intestinalis: Ascidians are invertebrate chordates closely related to vertebrates. Ascidians and vertebrates share the body plan including a dorsal neural tube, notochord, pharyngeal gill and endostyle/thyroid gland. Attention has focused on the cosmopolitan species, Ciona intestinalis, as a model organisms in recent years because its genome sequence has been decoded and accumulated EST/cDNA/protein resources. The recent genomic analyses have shown that this ascidians have the basic set of the genes for constructing chordate body plan with less redundancy of gene functions. Ciona intestinalis is an excellent organism to conduct genetic analyses for understanding gene functions. The inland culture system, transposon-based transgenesis and mutagenesis have been developed in this ascidian. By using this transgenic technologies, various transgenic lines have been accumulated which are splendid resources for studying genetic functions. This bioresource project involves collecting, maintaining and supplying wild types, inbred lines and transgenic/mutant lines.



Larval CNS cells expressing Kaede (left, magenta) remained after metamorphosis to form the adult CNS (right, magenta).

Key Strains/Studies

● ***swimming juvenile***	(Cellulose synthase mutant)
● ***no gill slit***	(*Hox1* mutant)
● **tissue-specific GFP marker strains**	(epidermis, notochord, muscle, neural tissues)
● **neuron Kaede strains**	(vGAT, vGLUT, vACHT>Kaede)

The metamorphosis of ascidians is characterized by the dynamic rearrangement of the body plan from the swimming larva to sessile adult. During metamorphosis, the larval central nervous system (CNS) is dramatically reorganized to form the adult nervous system. By using transgenic lines expressing KAEDE fluorescent protein in the nervous system, it was shown that the adult CNS is organized by the larval glial cells during ascidian metamorphosis (Horie et al., Nature 469: 525–528).

CORE FACILITY UPGRADING PROGRAM

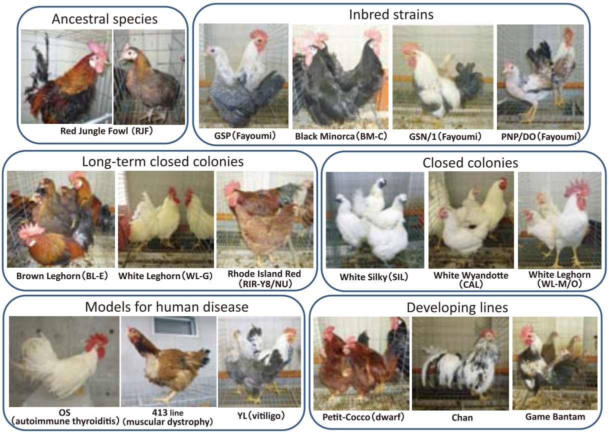
Chicken / Quail

Core Facility : Avian Bioscience Research Center, Nagoya University
Principal Investigator : Yoichi Matsuda FAX : +81-52-789-4099
Contact site : yoimatsu@agr.nagoya-u.ac.jp
URL : http://www.agr.nagoya-u.ac.jp/~nbrp/en/index.html

Overview

Chickens and quails are important model organisms in life sciences, which bridge the evolutionary gap between mammals and other vertebrates and serve as the main laboratory models for the ~ 9,600 extant avian species. Avian Bioscience Research Center (ABRC), Nagoya University contributes to advancement of avian science researches as the core facility of avian resources under NBRP.

The ABRC develops the stable system to maintain, preserve and distribute avian resources as chickens and quails. We also collect novel avian resources, develop them to the resources of high global standard under strict genetic control, and distribute them to the community of scientists. Furthermore, we construct the database of avian resources and enhance the database by adding science-based information obtained using the resources, which is widely open to the public via the homepage.



Chicken resources maintained in Nagoya University

Key Strains/Studies

The ABRC maintains and distributes more than 25 strains and/or lines of chickens including the red junglefowl (the wild ancestor of domesticated chickens), inbred strains, closed colonies, developing lines, and models for human diseases, and 17 closed colonies of Japanese quails including models for human diseases. Moreover, we collect and develop new avian resources that meet the research and social needs.

● GSN/1

This is a highly inbred strain originated from the Fayoumi chicken breed native to Egypt. This has been maintained as a closed colony for more than 30 years. Skin grafts are acceptable between different individuals within this strain. The genotyping of microsatellite DNA markers reveals that 40 loci are all fixed in the homozygous condition, indicating that this strain is very suitable for genetic analysis.

● RJF line

This line was originated from one pair of wild red junglefowls that were imported from Sumatra Is. (Indonesia), and mated with red junglefowls derived from Vietnam or Thailand, and then has been maintained as a closed colony. As the red junglefowl is ancestral species of domestic chickens and retains the wild-type characteristics, it can be used as a control for other domestic lines.

● WE

This is a Japanese quail line that has been maintained as a closed colony, which was introduced from Nippon Institute of Biological Science. This line is used for producing vaccine of Marek’s disease, toxicity assays of chemicals including pesticides, and toxicity tests in OECD and United States EPA. This line is designated as a standard line for reproductive toxicity tests of chemicals subject to the Chemical Substance Control Law from Ministry of the Environment, Japan. There is the highest demand for this line in Japanese quails.

WE line

CORE FACILITY UPGRADING PROGRAM Paramecium

Core Facility : Graduate School of Science and Technology for Innovation,
Yamaguchi University
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URL : http://nbrpcms.nig.ac.jp/paramecium/

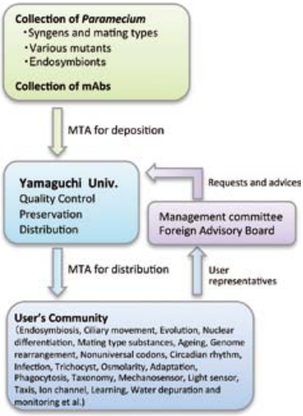
Overview

The ciliate *Paramecium* species are model organisms used for various researches on eukaryotes. Yamaguchi University is acting as the core facility for this resources in the world.

We collect, maintain and provide various species and their metadata (e.g. syngens and mating types) to researchers. We also provide strains used for genome or transcriptome studies, endosymbiont-bearing strains, and monoclonal antibodies developed for both the host and their endosymbionts.

Material Transfer Agreements (MTA) are used to protect the intellectual property rights of the developer of the strains and the antibodies. Yamaguchi University collaborates with overseas institutes to contribute for the advancement of the life science.

High-Quality *Paramecium* Resources

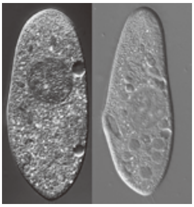


Key Strains/Studies

● *Paramecium caudatum* strain RB-1

Infection process of endonuclear symbiotic bacteria *Holospora* species that can distinguish nuclear envelopes of a vegetative macro- and a germinal micronucleus of the host *Paramecium caudatum* was found using this strain; i. e. escape from the host digestive vacuole, migration to the target nucleus by actin-based motility, distinguish two kinds of the host nuclear envelopes, penetration of the target nuclear envelope, and growth in the nucleus.

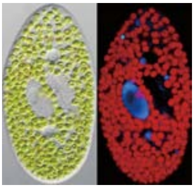
(Fujishima M. [Ed], Endosymbionts in *Paramecium*, Microbiology Monographs 12, Springer, 202–225, 2009)



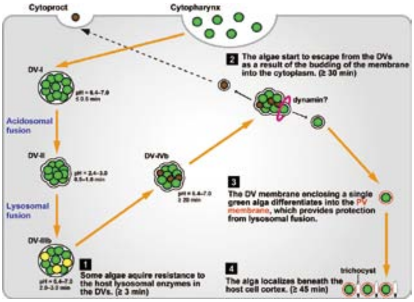
P. caudatum and *Holospora*.
Left, macronucleus specific *H. obtusa*-bearing cell. Right, micronucleus specific *H. undulata* bearing cell.

● *P. bursaria* strain Yad1g1N

Symbiotic *Chlorella*-free *P. bursaria* and the symbiotic algae retain the ability to grow without a partner. When the aposymbiotic host cells and the algae are mixed, they can reestablish the endosymbiosis synchronously. This system provides excellent opportunities for us to elucidate the secondary endosymbiosis leading to eukaryotic cell evolution. We found four important phenomena needed for the secondary endosymbiosis. (Updated from Kodama, Fujishima, Int. Rev. Mol. Cell Biol., 279, 33–77, 2010)



P. bursaria cell. Left, an alga-bearing cell. Right, DAPI fluorescence (blue) and autofluorescence of chlorophyll (red).



CORE FACILITY UPGRADING PROGRAM **Xenopus tropicalis**

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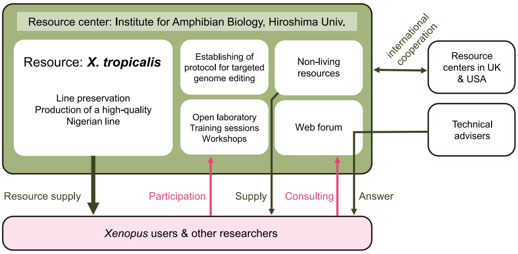
Overview

Since the Age of Enlightenment in 17th and 18th century Europe amphibians have long been important experimental animals. Approximately 10% of all Nobel Prizes in physiology or medicine have resulted from investigations using amphibians. The tropical clawed frog *Xenopus tropicalis* has only recently been introduced to the scientific community, and is still in the developmental stage as an experimental animal. This species can greatly simplify genetic studies because of its diploid genome and short generation time of approximately 6 months for males and 8 months for females. Moreover 79% of the genes bearing on human diseases are present as orthologues in *X. tropicalis*, and therefore *X. tropicalis* may be regarded as prime material for use in human disease research. At the Hiroshima University Institute for Amphibian Biology (IAB) we maintain 6 basal *X. tropicalis* populations consisting of 125 strains, which include about 10,000 individuals. Each year we supply more than 7,000 high quality individuals to various researchers and educators.

Using 60 highly polymorphic microsatellite markers, we investigated inbreeding ratios and genetic relationships between inbred strains at our facilities, as well as stocks from other institutions. Results show that successive reduction of heterozygosity has been successful in the genomes of IAB inbred strains, and that Nigerian A and NH strain individuals in particular are nearly identical clones. We performed mutual skin graft transplantations between 4th generation Nigerian H strain frogs, and the fact that the skin grafts have taken well suggests that these individuals have the same major histocompatibility complex haplotype. We produced knockout Golden strain frogs using CRISPR/Cas9 and found that 80 ~ 99% of the genes were targeted in F0 embryos, indicating that genome editing using high quality *X. tropicalis* makes it possible to easily and highly efficiently assess gene functions. Our recent study also shows that matrix-assisted laser desorption/ionization (MALDI)-mass spectrometry imaging (MSI) analysis is a powerful tool for identifying and visualizing metabolomic molecular markers in *X. tropicalis* tadpoles.



Resource center: Institute for Amphibian Biology, Hiroshima Univ.

Resource: ***X. tropicalis***

Line preservation
Production of a high-quality
Nigerian line

Establishing of
protocol for targeted
genome editing

Non-living
resources

Open laboratory
Training sessions
Workshops

Web forum

international
cooperation

Resource
centers in UK
& USA

Technical
advisers

Resource supply

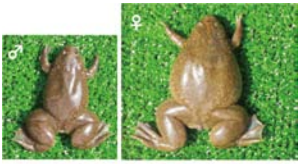
Participation

Supply

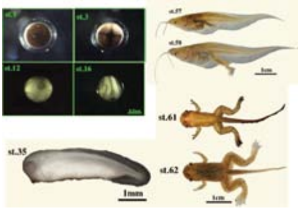
Consulting

Answer

Xenopus users & other researchers

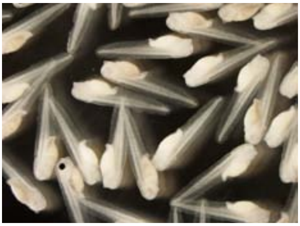


Xenopus tropicalis **(Nigerian H strain)**



A transgenic embryo expressing GFP in the developing ear and pronephros under the control of a cis-regulatory region of *Xenopus tropicalis pax8* gene.

Mutation in F0 Golden strain tadpoles using CRISPR/Cas system. The uppermost tadpole is the control.



First generation mutations.

Key Strains/Studies

- **Nigerian A line**
- **Golden line**
- **Nigerian H line**
- **Ivory Coast line**

The genome of the western clawed frog *Xenopus tropicalis* (Hellsten, Ogino, et al., Science, 2010)
Evolution of a tissue-specific silencer underlies divergence in the expression of *pax2* and *pax8* paralogues (Ochi, et al., Nature Communications 2012)
Inbreeding ratio and genetic relationships among strains of the Western Clawed Frog, *Xenopus tropicalis* (Igawa, et al., PLoS ONE 2015)
A streamlined workflow for rapid and efficient gene disruption by CRISPR-Cas9 in *Xenopus tropicalis* founders (Shigeta, et al., Genes Cells 2016)

● References

Ochi H, Tamai T, Nagano H, Kawaguchi A, Sudou N, Ogino H. (2012) Evolution of a tissue-specific silencer underlies divergence in the expression of *pax2* and *pax8* paralogues. Nat Commun. 3, Article number: 848.
Nakajima K, Yaoita Y. (2013) Comparison of TALEN scaffolds in *Xenopus tropicalis*. Biology Open, 2: 1364–1370.
Kurabayashi A, Kakehashi R, Tazawa I, Haramoto Y, Oshima T, Ito Y, Sumida M. (2014) Improved transport of the model amphibian, *Xenopus tropicalis*, and its viable temperature for transport. Curr Herpetol. 33: 75–87.
Yasuoka Y. *et al*. (2014) Occupancy of tissue-specific *cis*-regulatory modules by Otx2 and TLE/Groucho for embryonic head specification. Nat Commun. 5, 4322.

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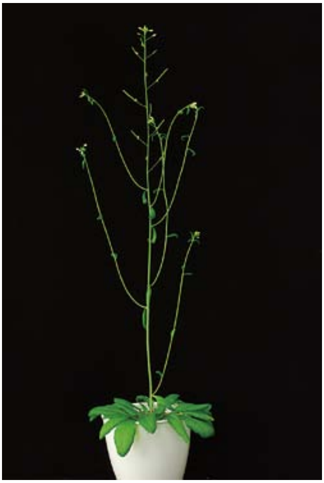
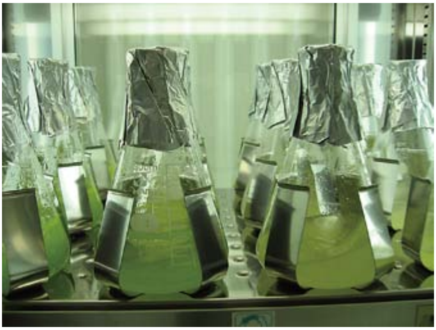
Overview

The small size and short life cycle of Arabidopsis make it an excellent model plant for experimental research. Ever since the sequencing of its genome was completed in December 2000, many international projects have been initiated around the world to identify the functions of its nearly 27,000 genes. Against this backdrop, there is a pressing need to develop a platform to support the application of the growing pool of Arabidopsis research findings to the betterment of society, such as the improvement of food production. The RIKEN BioResource Center (BRC) is contributing to efforts for tackling challenges related to food, the environment, and materials production not only by supplying researchers with seeds of gene-disrupted, wild-type, and other Arabidopsis stocks, but also by enhancing its resource databases, sharing technical information, and engaging in other productive efforts to aid plant research.

RIKEN BRC also supports research involving cultured plant cells and plant genes by storing and supplying model plant resources developed in Japan, and by disseminating information to research communities through coordination with other organizations participating in NBRP.



Arabidopsis at flowering stage



Arabidopsis T87 cell line

Key Strains/Studies

- **Arabidopsis transposon-tagged mutants (approx. 16,000 lines) and FOX mutants (approx. 18,000 lines)**
  These can be used as gene-disrupted mutants and over-expression mutants, respectively.
- **Arabidopsis full-length cDNA (RAFL) clones (approx. 250,000 clones, including about 21,000 completely sequenced clones)**
  These are standard resources used around the world.
- **Arabidopsis T87 cultured cells**
  These are the most frequently used Arabidopsis cells.

RIKEN BRC supplies its plant resources to more than 1,900 laboratories and research groups in Japan and abroad for use in a broad range of research. One example is the research group in Okayama University and RIKEN, which utilizes Arabidopsis full-length cDNA clone in research on isolation and characterization of Vitamin C transporter. Their report is published in Nature Communications.

CORE FACILITY UPGRADING PROGRAM **Rice**

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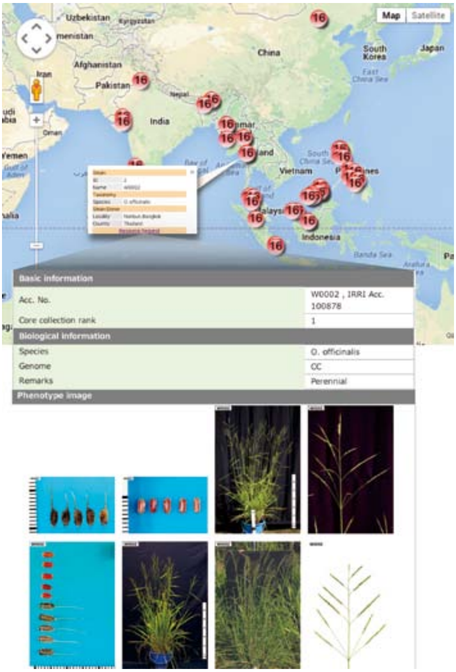
Overview

Japan has some of the world's most valuable collections of rice genetic resources. The NBRP-Rice collects, preserves, and provides a wide variety of genetic resources of wild and cultivated rice as well as valuable information of them for rice researchers. Specifically, the following efforts will be made aiming to meet different needs in current research areas of rice science.

(1) Preservation and provision of worldwide collection of wild rice species (23 species, 1,700 accessions). Characterization and reclassification of the wild accessions. Development of DNA markers to classify the wild rice species.
(2) Collection of experimental rice strains derived from wild species such as chromosome segment substitution lines (CSSLs) and monosomic alien addition lines.
(3) Collection of MNU-induced mutants derived from several varieties.
(4) Open laboratory supporting TILLING analysis to identify mutations in MNU-induced mutants
(5) Creation and management of ORYZABASE database for releasing rice resource and genomics information

The National Institute of Genetics functions as the core facility, managing all projects and handling the practical aspects of the wild species rice projects. Kyushu university is responsible for collecting mutants induced by the chemical (MNU) and CSSL series of wild species. Nagoya University shares responsibility for collecting other experimental strains of cultivated rice.



Basic information	
Acc. No.	W0002 , IRRI Acc. 100878
Core collection rank	1
Biological information	
Species	O. officinalis
Genome	CC
Remarks	Perennial
Phenotype image	

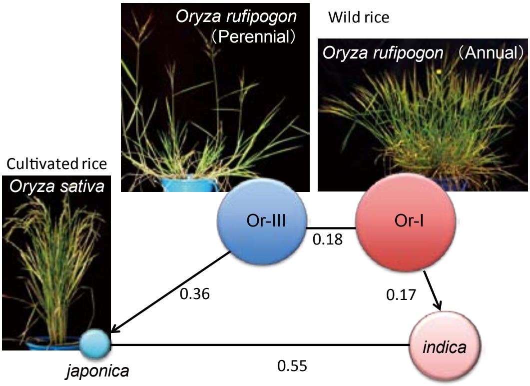
Locality map and details of wild rice accessions

Key Strains/Studies

- **Wild species rice strains**
  (approx. 6,800 strains)
- **MNU-induced mutant strains**
  (approx. 9,000 strains)
  (derived from Kinmaze, TC65, Kitaake, & Yukihikari)
- **Others including CSSLs of wild species**
  (approx. 4,800 strains)

Recent topics

NBRP-Rice preserves a wide range of wild rice species collected from around the world. Comparative genome analysis of these wild accessions and cultivars have revealed the origin of cultivation in the middle area of the Pearl River in southern China (Huang et al., Nature 490: 497–501, 2012).



Genetic diversity and population differentiation in cultivated rice and its wild progenitor

CORE FACILITY UPGRADING PROGRAM Wheat

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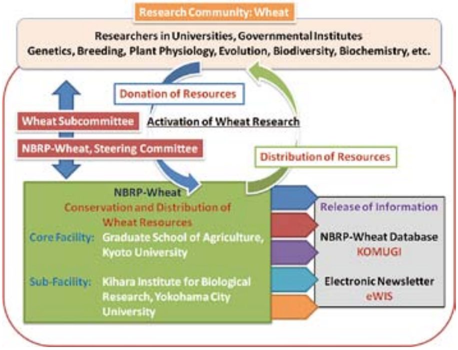
Overview

Wheat includes bread wheat, which is used to make bread and udon noodles, and macaroni wheat, which is used to make macaroni and other forms of pasta. The NBRP Wheat project stores and supplies wild species, landraces, and experimental strains of wheat and related species. It also collects and stores wild species and landraces that have not yet been archived, and stores and supplies EST and TAC clones of wheat. The project is implemented by the Graduate School of Agriculture, Kyoto University (core facility) and the Kihara Institute for Biological Research, Yokohama City University (sub-facility). The Wheat Subcommittee, which is organized by scientists who conduct wheat researches in Japan, supports the core team's work. The project's wheat resources can be requested online. The project members are also focusing on the characterization of DNA markers for use in gene isolation, and will release the resulting data in the NBRP section of the KOMUGI database.



NBRP-Wheat, Project Implementation System

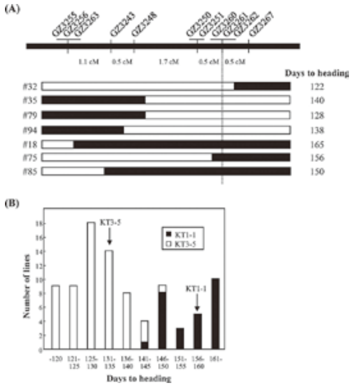
On 18 July 2014, the IWGSC published in the international journal Science a draft sequence of the bread wheat genome.

Key Strains/Studies

● Bread wheat aneuploids, ancestral species, landraces, full-length cDNA clones

Bread wheat is an allohexaploid species carrying three genomes (A, B, and D genomes) derived from diploid ancestral species. The main body of our genetic stocks is the aneuploids, the chromosome structural mutants, and the alloplasmic lines of a standard hexaploid wheat cultivar 'Chinese Spring'. Recent major advancement of wheat science is the exploration of genome sequence of the Chinese Spring wheat, which was has been led by the International Wheat Genome Sequencing Consortium (IWGSC). The draft sequences of the genes locating on each chromosome arm are currently available (IWGSC (2014) Science 345, DOI: 10.1126/science.1251788). In parallel, the BAC-based physical maps of wheat chromosomes are under construction and efforts are to be made to determine the sequences of the BAC contigs. Japanese team is in charge of the chromosome 6B (Kobayashi et al. (2015) BMC Genomics 16, DOI: 10.1186/s12864-015-1803-y). The genetic stocks of NBRP-Wheat are fully utilized in these genome projects.

The availability of genome-information of wheat and barley as the reference sequences has been changing the research environment in wheat. Gene identification by utilizing genome information is a feasible approach. The 'Early mutant' of a diploid what is known for its early heading nature for a long time and stored in a collection of induced mutants in NBRP-Wheat. Now we discovered that a deletion of a circadian clock gene *Phytoclock1* is responsible for the early heading phenotype (Mizuno et al. (2012) Genes Genet. Syst. 87, DOI: 10.1266/ggs.87.357).



A gene conferring early heading phenotype has been identified by utilizing the genome information of cereals.

CORE FACILITY UPGRADING PROGRAM **Barley**

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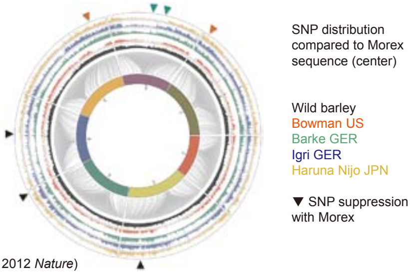
Overview

Barley is an important crop plant that serves many purposes, including malting, food, and animal feed. Diverse genetic resources of wild and cultivated barleys have been preserved, and a large range of experimental strains have been developed, particularly mutant strains. Recently, BAC libraries and extensive cDNA resources have been developed for genomic analysis of barley, which is a member of the tribe Triticeae and a model plant of cultivated wheat.

Okayama University's Institute of Plant Science and Resources, preserves and supplies barley strains collected or developed originally, a BAC library, and cDNA clones. The project has also developed and supplies filters for efficient selection of BAC clones, and has pooled DNA samples for PCR analysis. Moreover, the project distributes the world's first full-length cDNA clones of barley (developed in the NBRP genome analysis program), and publishes their nucleotide sequences, which contributed to estimate gene regions on the draft barley genome sequence published in 2012.

Diversity in barley spikes



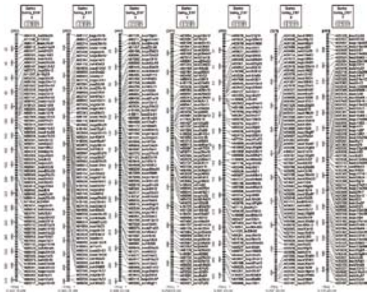
(IBSC 2012 *Nature*)

Haruna Nijo full length cDNA sequences are contributed to estimate gene regions on the draft barley genome sequence. 15 million SNPs are detected among haplotypes including Haruna Nijo sequenced by Okayama University.

Key Strains/Studies

- **Database publication and supply of approx. 6,000 strains, including Haruna Nijo, a standard strain for genomic analysis**
- **cDNA clones: 80,000 (Haruna Nijo, Akashinriki, ancestral wild strain H602)**
- **BAC library: 300,000 clones (Haruna Nijo)**

Barley is now being genetically mapped to analyze its agriculturally important genes, and the development of techniques for isolating and selecting those genes has a key challenge for researchers. The Okayama University project team has mapped to Haruna Nijo and wild barley segregating populations approx. 3,000 EST markers generated from 3' ESTs of the abovementioned resources. Upon mapping those markers to diploid wheat, the team found that barley and diploid wheat shared the same genomic structure, except for structural divergences in the fourth and fifth homoeologous chromosomes. This discovery shows that barley can be used as a diploid model of the Triticeae species, including wheat, barley and rye.



Genetic map of barley by 2,890 non-redundant 3'ESTs on cMAP platform
http://map.lab.nig.ac.jp:8085/cmap/

CORE FACILITY UPGRADING PROGRAM **Algae**

Core Facility : National Institute for Environmental Studies (NIES)
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http://mcc.nies.go.jp/ (Microalgae; NIES)
http://www.research.kobe-u.ac.jp/rcis-ku-macc/ (Macroalgae; Kobe University)

Overview

Algal bioresources include evolutionarily diverse organisms, such as prokaryotic cyanobacteria, photosynthetic eukaryotes except for the so-called land plants, as well as non-photosynthetic relatives (protozoa) (Fig. 1 & 2). These algae are also used for a wide-rang of researches such as life sciences, evolutionary and environmental studies. Selected algae are designated as model organisms, which are used for specific researches (for example photosynthesis and sexual reproduction). The National Institute for Environmental Studies (core facility: collection, preservation and supply of microalgae, collection acronym: NIES), the Kobe University (sub-facility: collection, preservation and supply of macroalgae, collection acronym: KU-MACC), the University of Tsukuba (sub-facility: securing taxonomic consistency and dissemination of taxonomic information of algal bioresources), and the Hokkaido University (sub-facility: backup of algal bioresouces) aim to be culture collections with the highest quality algal bioresources by collecting important species, adding valuable information to the strains maintained, and creating a quality control system.

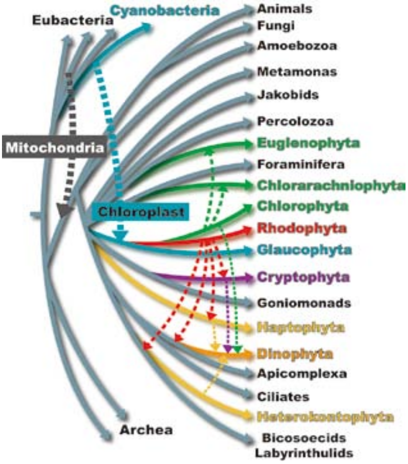

Fig. 1. Tree of life
Various phylogenetic groups are found in the eukaryotes. Colored branches indicate the algal groups, and the broken lines designate the organisms from which the chloroplast originated. The chloroplasts of Chlorophyta, Rhodophyta, and Glaucophyta are all originated from cyanobacteria. In contrast, the chloroplasts of the other algal groups are originated from particular chlorophyte or rhodophyte.

Key Strains/Studies

A diverse collection of algae including 564 genera, 1,060 species and 3,702 strains are available for the purpose of education, research and development, and have been used for a diverse range of researches as follows:

- **Model organisms (photosynthesis, sexual reproduction, cell division, etc.):** *Thermosynechococcus elongatus* (NIES-2133/BP-1), *Cyanidioschyzon merolae* (NIES-3377/10D), *Chlamydomonas reinhardtii* (NIES-2235/C-9), *Ectocarpus siliculosus* (KU-1371).
- **Evolutionarily important species:** *Mesostigma viride* (NIES-296), charophyte (NIES-1601).
- **Harmful algal species:** *Microcystis aeruginosa* (NIES-44), *Chattonella marina* (NIES-3).
- **Ecologically significant species:** *Prochlorococcus marinus* (NIES-2086) and *Micromonas pusilla* (NIES-1411) as major primary producers.
- **Commercially useful strains:** *Botryococcus braunii* (NIES-836), *Porphyridium* sp. (NIES-1035), *Chlorella vulgaris* (NIES-227).

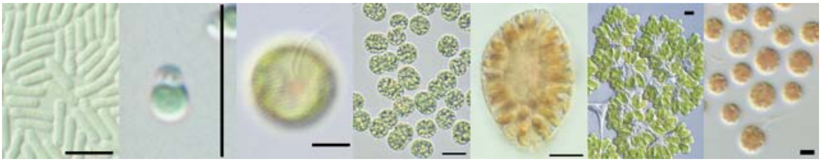

Fig. 2. Light micrographs of various algal resources.
From left side. *Thermosynechococcus* (NIES-2133), *Cyanidioschyzon* (NIES-3377), *Mesostigma* (NIES-296), *Microcystis* (NIES-44), *Chattonella* (NIES-3), *Botryococcus* (NIES-836), *Porphyridium* (NIES-1035).
Scale bar = 10 μm.

CORE FACILITY UPGRADING PROGRAM Chrysanthemum

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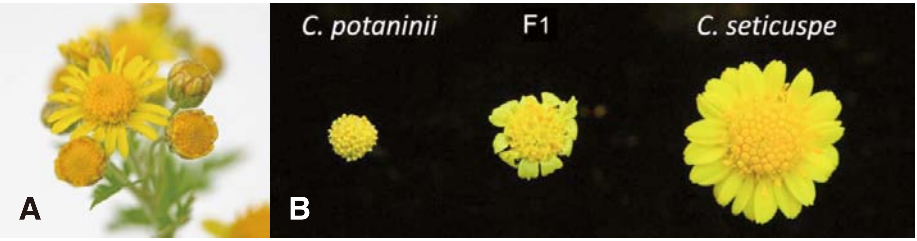
Overview

Asteraceae, which consists of more than 1,600 genera and 23,000 species, is one of the largest families in angiosperm and distributed worldwide except Antarctica. Among its members, the genus *Chrysanthemum* has evolved in the Eurasian continent and its surrounding regions and contains autoploid species from diploid to decaploid endemic to Japan. The cultivated chrysanthemum is unsuitable for genetic analysis due to its autohexapolidy. The diploid *Chrysanthemum* species *C. seticuspe* is anticipated to be a model species to elucidate various properties in cultivated chrysanthemum at molecular level because its flowering and inflorescence morphology properties resemble those of the cultivated chrysanthemum and its transformation method has been established. Furthermore, because hybridization between *Chrysanthemum* species is easy, *C. seticuspe* is expected as a platform for molecular analysis of genetic diversity in the genus *Chrysanthemum*. NBRP Chrysanthemum resources include other species with various morphology and characteristics. For example, *Xanthisma gracile* (synonym: *Haplopappus gracilis*) is known as a species with the least chromosome number (2n=4) in plants. Some species contain various useful secondary metabolites such as the commercially important antimaralia element. These bioresources are maintained in the field, pots, or *in vitro* to be provided to researchers and breeders.

Various *Chrysanthemum* plants and related species.

Key Strains/Studies



***C. seticuspe* (A) and the 'flowers' of F_1 hybrid between *C. seticuspe* and *C. potaninii* (B).**

The 'flower' of the genus *Chrysanthemum* is a florescence formed with many florets, called capitulum. Typically, the florets of the outer layer have long petals (ray florets) and those of inner layer have short tube-like petals (tubular florets). *C. potaninii* has only tubular florets but its F_1 hybrid with *C. seticuspe* has ray florets in the outer layer.

Morning glory

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URL : http://mg.biology.kyushu-u.ac.jp/

Overview

The Japanese morning glory (*Ipomoea nil* or *Pharbitis nil*) is a bioresource that was developed in Japan along with more than a century of knowledge amassed from its applications in genetics, physiology, natural product chemistry, and other disciplines. It has several strong advantages that make it useful for research in various areas of plant science, such as its highly homogeneous genome, which is the result of its high selfing rate and its restricted origin, as well as its diverse array of mutant variations related to flower color and morphology that are induced by its highly active transposons. It is expected to grow in importance in the years ahead for its usefulness in applied research, including its use in ornamental horticulture, and its use as a model organism for the sweet potato, which is a member of the same genus. Under the NBRP Morning Glory project, Kyushu University (core organization) and the National Institute of Basic Biology (sub-organization) are building up a repository for the collection, preservation, and supply of mutant strains, DNA clones, mutated gene information, and other resources. It is expected that the Japanese morning glory will become one of Japan's leading bioresources.

Key Strains/Studies

● Tokyo Kokei Standard (TKS)

Dr. Yo Takenaka of the National Institute of Genetics selected a strain considered close to wild-type Japanese morning glories, and developed into a standard strain through repeated selfing. This strain was used to construct the DNA libraries used in this project. The transposition rate of *Tpn*-transposons is suppressed.

● Violet

This is widely used as a standard strain for plant physiology research. It bears flower buds that are highly sensitive to short-day conditions. It includes the mutations *magenta* (*mg*) and *dragonfly* (*dg*).

Anthocyanins, a class of flavonoids, color many flowers. The depth of flower color is determined by the amount of anthocyanin. A new mutant with reduced amounts of anthocyanin whose flowers were paler in color was isolated and characterized. As a result, a protein that increases production efficiency of flavonoids including anthocyanin, leading to a darker color in the flowers, was found and named Enhancer of Flavonoid Production (EFP). It is expected that EFP can be applied to developing varieties of flowers and fruits having new value by modifying the content of flavonoids.
(Morita et al., Plant J. 78, 294–304, 2014)

An *efp* mutant with defects in anthocyanin accumulation (left). A wild-type (right).

CORE FACILITY UPGRADING PROGRAM **Lotus / Glycine**

Core Facility : Faculty of Agriculture, University of Miyazaki
Principal Investigator : Ryo Akashi FAX : +81-985-58-7257
Contact site : legume@brc.miyazaki-u.ac.jp
URL : https://www.legumebase.brc.miyazaki-u.ac.jp/

Overview

Legumes are cultivated and used as a vital source of protein from the tropics to the temperate zones, and they are also an important resource for research on such topics as the accumulation of secondary metabolites and symbiosis with microorganisms.

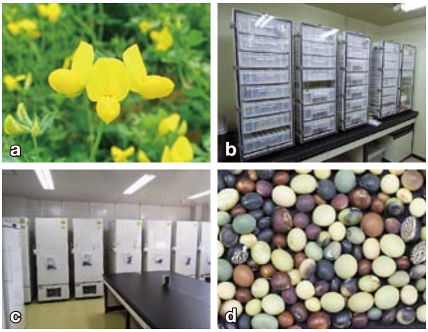
Lotus japonicus (Japanese trefoil) is a perennial legume that grows naturally throughout many parts of Japan. It is widely used as a model plant for legumes because of such advantages as its small genome size, short life cycle, etc. Soybeans (*Glycine max*) have been grown as an important crop since ancient times, and a large amount of basic research has been conducted on them over the years. The *L. japonicus* resources and information that have been collected and developed are indispensable for improving the efficiency of a great variety of soybean-related research, including studies aimed at increasing soybean yields based on symbiosis with *Rhizobium*, and at enhancing the nutritional functions of soybeans.

This project seeks to construct a platform for legume research by collecting, preserving, and supplying a broad set of resources that can support both basic research involving *L. japonicus* and applied research involving soybean.

Key Strains/Studies

This project has preserved approximately 4,000 plant resources. The *L. japonicus* collection is mainly 3 experimental strains and approx. 200 wild accessions, and a database is being built up based on this collection. The project has also preserved a variety of soybean strains, including wild strains and recombinant inbred lines. This project has collected more than 220,000 BAC, TAC and cDNA clones derived from *L. japonicus* Miyakojima MG-20 and soybean full-length cDNA clones derived from Norin No. 2.

The experimental strains, Gifu B-129 and Miyakojima MG-20, which are mainly used in research pertaining to root nodules (where the plant has a symbiotic relationship with microorganisms) and mycorrhiza formation. A number of important genes have been isolated through such research.



a: Flower of *Lotus japonicus*, b: Seed stock room, c: DNA clone stock room, d: Various soybean seeds.

Resource type preserved in this project.

Lotus resource
Wild accession
Experimental strain (Gifu B-129, Miyakojima MG-20, *Lotus burttii* B-303)
RIL (Gifu B-129 x Miyakojima MG-20)
RIL G x LB (Gifu B-129 x *L. burttii* B-303)
RIL F x G (*Lotus filicaulis* B-37 x Gifu B-129)
EMS mutant (Miyakojima MG-20)
Root culture; superroot (*Lotus corniculatus*)
EMS M2 bulked seed (Miyakojima MG-20)
LORE1 tag line (Gifu B-129)
Mesorhizobium loti STM mutant
TAC clone (Miyakojima MG-20)
BAC clone (Miyakojima MG-20)
cDNA clone (Gifu B-129 and Miyakojima MG-20)
Full-length cDNA clone (Gifu B-129 and Miyakojima MG-20)
Transformation vector
Glycine resource
Wild accession (*Glycine soja*)
Cultivar (*Glycine max*)
RIL (Misuzudaizu x Moshidou Gong 503)
RIL (TK780 x B01176)
Edamame (vegetable soybean)
Mutant (Fatty acid, Symbiosis, Flower color)
EMS M2 bulked seed (cv. Fukuyutaka)
Full-length cDNA clone (cv. Norin 2)
BAC clone (*Bradyrhyzobium japonicum*)
Cosmid clone (*B. japonicum*)

CORE FACILITY UPGRADING PROGRAM Tomato

Core Facility : Gene Research Center, Faculty of Life and Environmental Sciences,
University of Tsukuba
Principal Investigator : Hiroshi Ezura FAX : +81-29-853-7734
Contact site : ezura@gene.tsukuba.ac.jp
URL : http://tomato.nbrp.jp/

Overview

Tomato is an excellent model plant for studying fruit development. Tomato is considered as a model species of Solanaceae, and its genome sequence has determined by international consortium. In order to fully utilize the genome information, the core facility, University of Tsukuba, and a sub-facility, Osaka Prefecture University, launched on the tomato resource development program within the framework of the National BioResource Project. The University of Tsukuba and Osaka Prefecture University take charge of developing individual-and DNA-level resources, respectively.

Key Strains/Studies

The University of Tsukuba will maintain and distribute 1) tomato wild varieties, 2) tomato transgenic lines, 3) M3 seeds of approximately 14,000 Micro-Tom mutant lines (Figures 1 and 2), comprised of 4,909 gamma-ray irradiated and 9,000 EMS treated lines. These materials are available at : http://tomato.nbrp.jp/ upon request. Osaka Prefecture University maintains 292,336 full-length cDNA clones developed from Micro-Tom fruits, leaves and roots, 314,496 tomato BAC clones, and 32 tomato promoter clones. The full-length cDNA clones are available upon request. As of March 2016, University of Tsukuba has created a database of the phenotype information in 96 experimental lines and 2,290 mutant lines that were selected during the process of M3 seed propagation. This database, called Tomato Mutant Archives (TOMATOMA), was developed with assistance from the Genetic Resource Center at the National Institute of Genetics. Information of full-length cDNA sequences of 13,227 clones and 5’-end sequences of 89,872 clones are available at full-length cDNA database KaFTom (http://www.pgb.kazusa.or.jp/kaftom/), EST database MiBASE (http://www.pgb.kazusa.or.jp/mibase/), and integrated omics database TOMATOMICS (http://bioinf.mind.meiji.ac.jp/tomatomics/) both of which were developed by collaboration with Meiji University and Kazusa DNA Research Institute. In addition, 93,682 Micro-Tom BAC end-sequences have been determined and their information is available at DNA database banks.

Figure 1.
A model cultivar, Micro-Tom

Figure 2.
Micro-Tom mutants

Cellular slime molds

Core Facility : Faculty of Life and Environmental Sciences, University of Tsukuba
Principal Investigator : Hidekazu Kuwayama FAX : +81-29-853-4664
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URL : http://nenkin.nbrp.jp/

Overview

Cellular slime molds are unicellular amoebae that feed on bacteria and proliferate by fission. One of their key features is that when they are placed under starvation stress, they aggregate to form multicellular structures that develop into fruiting bodies composed of spore balls and supporting stalks. They are used as a model organism for research on development, cell division/motility, mathematical biology, and so forth. They are also employed as a resource for medical research because of several useful features, such as: they can serve as hosts for pathogenic microorganisms and they serve novel physiological active substances .

This project is being implemented by the University of Tsukuba, the National Institute of Advanced Industrial Science and Technology (AIST) and RIKEN Q-Bic with the goals of developing a world-class collection of high-quality cellular slime mold resources and providing a broad range of services to answer the needs of researchers in Japan. As part of this endeavor, the project members engage in the following activities:

(1) Collection, preservation, and supply of various strains
(2) Collection, preservation, and supply of genes and vectors
(3) Hosting of training courses for new users

Cellular slime molds are easy to culture, and most molecular biology techniques can be applied to them. Moreover, they are haploid for most of their life cycle. Even laboratories with no prior experience in working with cellular slime molds will find them readily usable as asecondary resource.

Key Strains/Studies

Dictyostelium discoideum
NC4, KAX3, AX2, AX4, and V12 strains, and genetically manipulated strains
Other strains available:
Dictyostelium mucoroides, Dictyostelium purpureum, Polysphondylium pallidum, Acytostelium subglobosum, and more
Clones:
D. discoideum cDNA clones which cover more than 60% of total genes (about 12,500) and expression vectors with various tags
A. subglobosum cDNA and genomic clones

D. discoideum AX4 is the strain that was used to analyze that species' genome and EST. KAX3 and AX2 are widely used in molecular biology analysis, and many development-related genes have been identified. More than 2,000 of the cDNA clones have been supplied by the University of Tsukuba to a diverse assortment of research projects around the world. The entire gene sets were used for microarray analysis, and studies using these sets have shed light on such topics as the changes in gene expression patterns that accompany transformation to a multicellular structure, and the infection mechanism of *Legionella* bacteria, which can cause pneumonia. Furthermore, collected new strains are under investigation for novel physiological active substances.

CORE FACILITY UPGRADING PROGRAM Pathogenic microorganisms

Core Facility : Medical Mycology Research Center, Chiba University
Principal Investigator : Takashi Yaguchi FAX : +81-43-226-2486
Contact site : yaguchi0801@gmail.com URL : http://www.pf.chiba-u.ac.jp
Takayuki Ezaki, Gifu University Contact site: tezaki@gifu-u.ac.jp
Tetsuya Iida, Osaka University Contact site: iida@biken.osaka-u.ac.jp
Kenji Hirayama, Nagasaki University Contact site: hiraken@nagasaki-u.ac.jp

Overview

This project is carried out by Chiba University's Medical Mycology Research Center (pathogenic fungi/actinomycetes), Osaka University's Research Institute for Microbial Diseases (pathogenic bacteria), Gifu University's Graduate School of Medicine (pathogenic bacteria), and Nagasaki University's Institute of Tropical Medicine (pathogenic protozoa). Together, they cooperate in various efforts to support education and research pertaining to infectious diseases and pathogens. Specifically, they are developing a system for collection, preservation, and distribution of pathogenic microorganisms, and they supply reliable strains of pathogenic microorganisms that are backed by high-level information. They also contribute to efforts aimed at conquering infectious diseases.

Since the project deals with pathogenic microorganisms, there are some resources that have restricted database access or require clients to furnish details of their organization. Those interested in ordering resources are asked to consult with a project representative.

Key Strains/Studies

- **Pathogenic fungi, bacteria, protozoa:** All species of highly pathogenic exogenous fungi (including type 3 pathogens), more than 90% of other pathogenic fungi species, standard strains of pathogenic actinomycetes (mainly *Nocardia*), more than 80% of human-pathogenic bacteria species, and human-infecting protozoa (information of strains preserved by other institutions).

Research contributions: The project's resources have been used as, among other applications: (1) strains for creation of DNA chips for identification of pathogenic microorganisms, (2) strains for full genomic analysis, (3) standard strains for proposing new species, (4) comparative strains for drug susceptibility testing of clinical isolates, (5) resources for student experiments, and (6) resources for drug development by pharmaceutical companies.

CORE FACILITY UPGRADING PROGRAM General Microbes

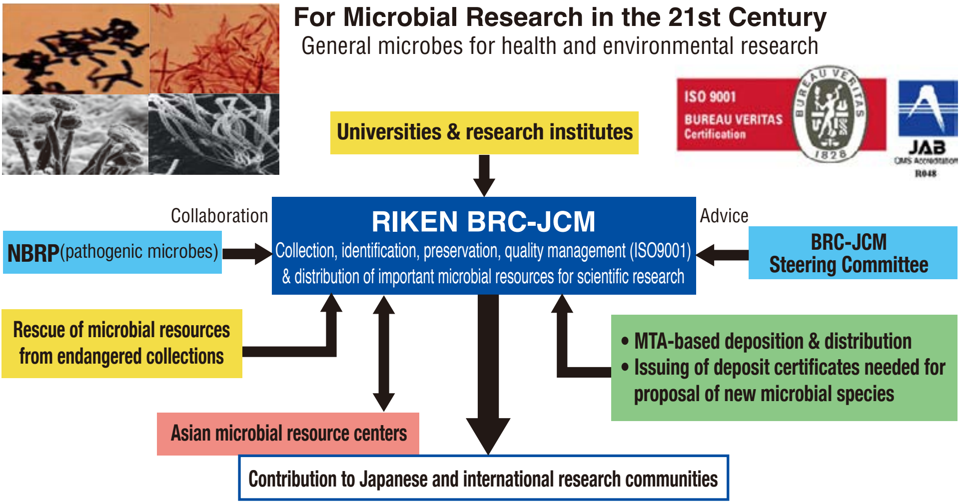
Core Facility : Microbe Division/Japan Collection of Microorganisms (JCM),
RIKEN BioResource Center
Principal Investigator : Moriya Ohkuma FAX : +81-29-836-9561
Contact site : inquiry@jcm.riken.jp
URL : http://jcm.brc.riken.jp

Overview

The Japan Collection of Microorganisms (JCM) has contributed to bioscience and biotechnology as one of the leading culture collections in the world since established in 1981. A wide variety of microbes that are particularly relevant to human health science and environmental science are strategically collected, preserved, and distributed. Approximately 25,000 strains comprising of type strains and their related strains of aerobic and anaerobic bacteria, archea, yeasts, and fungi are collected and preserved, and in each year, more than 4,000 strains are distributed to researchers. Distribution of microbial genome DNA has been started through the collaboration with the Gene Engineering Division of RIKEN-BRC. JCM is certified by ISO9001:2008, an international standard of quality management system. JCM has accepted endangered culture collections such as the IAM collection of The University of Tokyo. JCM continually updates the latest information of the JCM strains on the online catalogue. Efforts have also been made to exploit new microbial resources and to develop technologies for handling extremophiles and uncultured microbes.

Key Strains/Studies

- **Lactic acid bacteria, anaerobes, aerobes, actinomyces, archaea, yeasts, fungi, and more**



For Microbial Research in the 21st Century

General microbes for health and environmental research

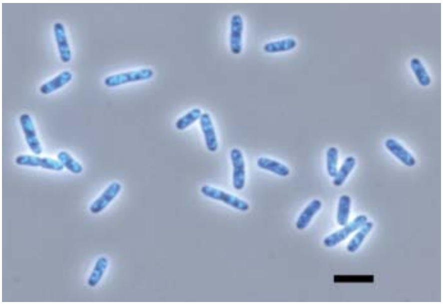
Prokaryotes (E. coli, B. subtilis)

Core Facility : Genetic Resource Center, National Institute of Genetics
Principal Investigator : Hironori Niki FAX : +81-55-981-6826
Contact site : genkaku@nig.ac.jp
E. coli URL: http://www.shigen.nig.ac.jp/ecoli/strain/
B. subtilis URL: http://www.shigen.nig.ac.jp/bsub/

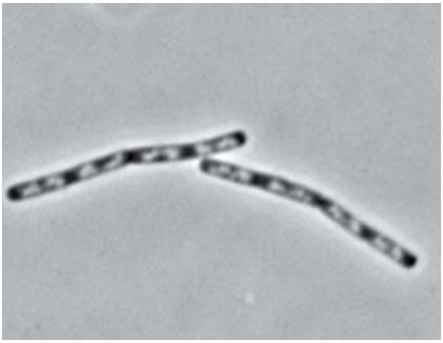
Overview

The National Institute of Genetics collects and preserves *Escherichia coli* and *Bacillus subtilis* resources developed in Japan, and supplies them to institutions that engage in basic research. Resource orders are handled through the webpage for each species, so users can request delivery of their desired strains by filling out the necessary information online. Delivery is made to domestic addresses within one week of ordering. Fees of strains and DNA are shown in tour web home page. Shipping fees are usually born by the user. In addition, in order to receive resources, the leader of the research project must sign a materials transfer agreement (MTA) with the National Institute of Genetics. For certain resources that are genetically modified organisms, users also need to obtain a permit for handling the organism.

The National Institute of Genetics acts as the core organization for this NBRP project, and sub-organization Kyushu University handles the preservation of resources in order to ensure their safe storage.



Escherichia coli



Bacillus subtilis

Key Strains/Studies

The resources distributed by this project are all nonpathogenic strains. *E. coli* strains are derived from strain K12, and *B. subtilis* strains are derived from strain 168. The project's collection consists of *E. coli* gene clones with GFP- or His-tag on expression vectors. The *E. coli* resources are largely divided into the following three groups.

- **Mutant *E. coli* strain resources** (exhaustive collection of gene-knockout mutants, transposon-insertion mutants, etc.)
- **Cloned genetic resources** (GFP-tagged and untagged ASKA clones)
- **Cloning vector** (465 types) and host resources

The *B. subtilis* resource collection consists of nearly 2,500 gene-disrupted strains created mainly by the Ogasawara Laboratory of the Nara Institute of Science and Technology (Kobayashi et al., 2003). The disrupted genes of all strains are being checked by PCR. The strains that pass this quality control check have been released and distributed as they become available. The host stain for the seamless cloning is now available.

CORE FACILITY UPGRADING PROGRAM Yeast

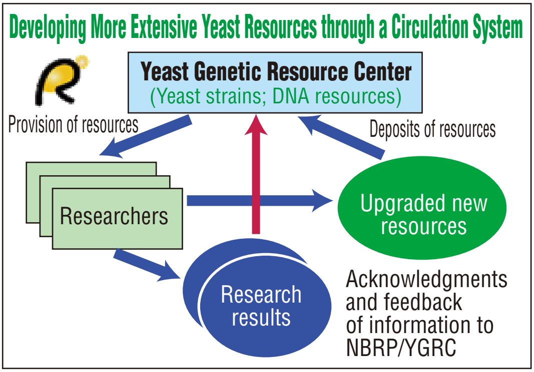
Core Facility : Graduate School of Science, Osaka City University
Principal Investigator : Taro Nakamura FAX : +81-6-6605-2576
Contact site : nbrpombe@sci.osaka-cu.ac.jp (Taro Nakamura)
bygrc@bio.eng.osaka-u.ac.jp (Minetaka Sugiyama)
URL : http://yeast.lab.nig.ac.jp/nig/

Overview

Yeast is an important eukaryotic model organism. This is especially true of the fission yeast *Schizosaccharomyces pombe* and the budding yeast *Saccharomyces cerevisiae*, which are making significant contributions to research in a variety of areas within the life sciences.

Upgrading bioresources and expediting their distribution are essential to successful research. Through phases 1 and 2 of the NBRP, the Yeast Genetic Resource Center (YGRC) has become one of the top international yeast resource centers. In phase 3 of the NBRP, YGRC will aim to possess various high-quality resources, such as genome-wide and highly-demanded resources.

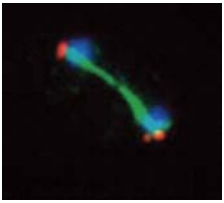
The project will be managed by the Graduate School of Science, Osaka City University (fission yeast) and the Graduate School of Engineering, Osaka University (budding yeast). The Center for Gene Science, Hiroshima University, handles the preservation of “Back-up” resources in order to ensure their safe storage. The “Yeast Genetic Resource Center Steering Committee” is functioning effectively as a contact point between the project and resource users.



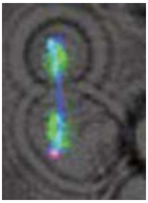
Developing More Extensive Yeast Resources through a Circulation System

Fission yeast

Budding yeast



Visualisation of the nuclear division (fission yeast)



Visualisation of the nuclear division (budding yeast)

Key Strains/Studies

- **Fission yeast** (approx. 18,000 strains; approx. 100,000 DNA clones)
  Mutant strains related to sexual reproduction and mitotic division; GFP-fusion gene library; sequenced full-length cDNA and genome DNA clone sets; various cDNA and genome libraries; temperature-sensitive mutant set; cold-sensitive mutant set
- **Budding yeast** (approx. 27,000 strains; approx. 5,600 DNA clones)
  Mutant strains related to cell cycle and cell wall sysnthesis; auxin-induced degron resources; double-disruptant-set of protein phosphatase genes; double gene disruptants of protein phosphatase and protein kinase; gTOW6000 strains; septin-related resources; ribosome-related resources; DNA barcode strains.

1. Analyses of intracellular protein localizations and protein dynamics have been conducted using GFP-fusion genes.
2. An analysis using full-length cDNA clone sets to clarify the yeast transcriptome is well under way and the results look very promising.

DNA material

Core Facility : Gene Engineering Division, RIKEN BioResource Center
Principal Investigator : Takehide Murata FAX : +81-29-836-9120
Contact site : dnabank@brc.riken.jp
URL : http://dna.brc.riken.jp/index.html

Overview

Genetic experimental materials, such as plasmid and viral vectors, genomic and cDNA clones, have become one of the most important and fundamental research tools for life sciences. Genetic materials are now widely utilized in numerous fields of life sciences, not only in basic researches such as analyses of gene function and control mechanisms of gene expression but also in applied researches such as development of novel diagnostic and therapeutic methods, drug discovery and material production. The Gene Engineering Division of RIKEN BioResource Center (BRC) has been engaging in the collection, preservation, quality control and distribution of genetic materials developed mainly in Japan by individual scientists and by various national projects.

Recently, genetic materials can be prepared easily by means of, for example, polymerase chain reaction. However, such materials often contain accidental mutations and produce dubious experimental results. To provide domestic and international scientific community with genetic materials of the highest quality and reproducibility, the Gene Engineering Division performs rigorous quality control by testing growth and propagation, restriction enzyme mapping and nucleotide sequencing of clones. Relevant information such as characteristics and methodologies is provided via the web site of the RIKEN BRC. For the best use of genetic resources, training courses of advanced technologies are also given.

The Material Transfer Agreement is used for each transfer of genetic materials to protect the intellectual property rights of developers and to define the responsibility of users. We have opened a path of the academic use of genetic materials produced by using advanced research tools owned by commercial entities. Deposition of genetic materials in the RIKEN BRC frees researchers from time consuming preparation and distribution of materials to fellow researchers. Furthermore, deposition increases chance of collaboration and citation of research papers. Your deposition of genetic resources in RIKEN BRC Gene Engineering Division and use of these resources are most appreciated.

Key Strains/Studies

● Deposited clones from researchers around the world

[Genomic (BAC) and cDNA clones of human, model animals and microorganisms]

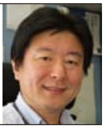
Numerous clones developed by individual researchers, and large sets of genetic materials from the core facilities of NBRP and other National Projects are available. The KEGG (Kyoto Encyclopedia of Genes and Genomes) database of pathways and orthologs among human, mouse and fission yeast are now linked to our clones and can be searched by users.

● Ready for use genetic materials

[Recombinant adenoviruses, promoter and reporter constructs, and expression vectors]

Users can readily perform their experiments using our genetic materials without re-constructing in an expression vector. For example, luciferase reporters to study signal cascades of Hedgehog, Notch, and Wnt/β-catenin as well as promoter constructs from 300 human genes are available.

CORE FACILITY UPGRADING PROGRAM Human and animal cells



Core Facility : Cell Engineering Division, RIKEN BioResource Center
Principal Investigator : Yukio Nakamura FAX : +81-29-836-9130
Contact site : cellqa@brc.riken.jp (Regarding materials and methods)
cellbank@brc.riken.jp (Regarding deposit or provision)
URL : http://www.brc.riken.jp/lab/cell/english/

Overview

Cell lines are a relatively easy-to-use research material, which can be used by anyone at any time and any place. They are also an extremely convenient research material because they can multiply virtually forever in a test tube. This convenience, however, is a double-edged sword. Researchers who are just starting out often become involved in cell culturing and this frequently gives rise to problems such as “cell line mix ups,” “mycoplasma contamination,” and so forth. When specimens affected by these problems end up being used in research, it results in work that is inaccurate and unreproducible. Scientifically valid conclusions thus cannot be obtained. In our division, we have developed a highly reliable system that provides cell specimens which are confirmed to be free of these problems. We invite all researchers considering new research projects to obtain cells from our ISO 9001 certified facility. (Fig. 1).

Fig. 1

Key Strains/Studies

(1) General Cell Lines

- Human Cancer Cell Lines, Human Primary Cells
- Animal Cell Lines (Mouse, Rat etc.)

(2) Cells for Genome Research

- Cells of Healthy Japanese
- Sonoda-Tajima Collection (Various Racial and Ethnic Background)
- Cells of Patients (Werner Syndrome, Breast Cancer etc.)

(3) Stem Cell Bank

- Human Somatic Stem Cells (Umbilical Cord Blood, Mesenchymal Stem Cells)
- Embryonic Stem Cells (Human, Common Marmoset, Mouse [Fig. 2])
- Induced Pluripotent Stem Cells (Human, Mouse, Rabbit)
- Disease-specific iPS cells (Human)

Fig. 2

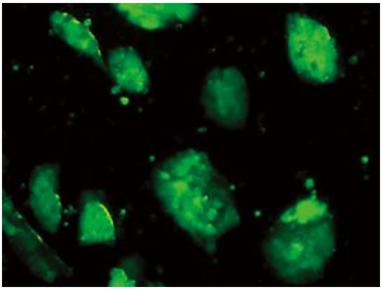
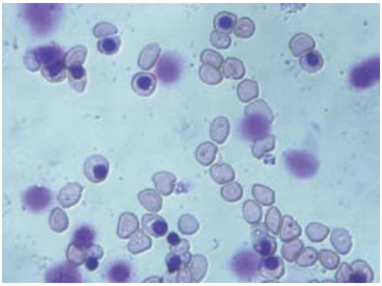


Fig. 3



Mature red blood cells produced in vitro from hematopoietic stem cells present in human umbilical cord blood samples.

There are already an enormous number of studies that have been completed using the division’s cultured cells. Please refer to the information that is available at the following URL: http://www.brc.riken.jp/lab/cell/english/

Using hematopoietic stem cells from human cord blood, for example, investigators have succeeded in developing a technique that produces large amounts of red blood cells in test tubes. Through use of this method, it becomes possible to produce nearly one million red blood cells from a single hematopoietic stem cell, without using other cells (feeder cells). Moreover, the red blood cells that are produced are virtually all denucleated red blood cells (Fig. 3).

Cord blood stem cells for research

Core Facility : Department of Cell Processing and Transfusion, Research Hospital,
The Institute of Medical Science University of Tokyo (IMSUT)
Principal Investigator : Tokiko Nagamura-Inoue FAX : +81-29-836-9130
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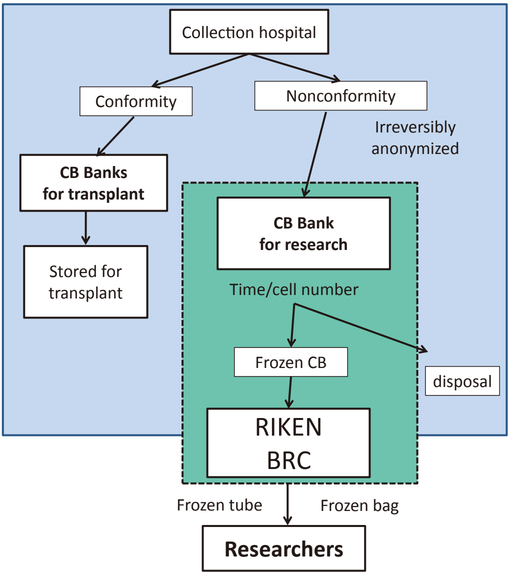
URL : http://www.brc.riken.jp/lab/cell/hcb

Overview

Human cord blood cells (CBCs) are known to contain the most premature hematopoietic stem cells as well as mesenchymal stem cells. CBCs have been clinically used as the source of hematopoietic stem cell transplantation for severe hematologic diseases like leukemia, and they are now widely used for research purposes in the medical and biological studies of regenerative medicine, drug development, epidemiology, infection, genetics and environmental studies.

This project provides frozen CBCs for research use, to researchers through the RIKEN BRC. The research CBs are collected with written consent, but no longer suitable for transplant purpose in clinical public CB banks, processed and cryopreserved into frozen CBCs in CB bank (IMSUT) for research and transferred to RIKEN BRC.

Flow chart of cord blood



Key Strains/Studies

● Mononuclear cells (CBF)

- preparation: 2ml in tubes (4 in one set) or Frozen Bag
- cell numbers > 1×10^7/tube or > 1×10^8/bag

※can choose sets from the same donor or different donors

- method: Ficoll sedimentation
- neutrophil less than 20% at freezing

● Nucleated cells (HCB)

- preparation: plastic bags
- cell numbers > 3×10^8/bag
- method: HES sedimentation

● CD34positive cells (C34)

- preparation: 2ml in tubes
- cell numbers > 1×10^5/tube
- method: immunomagnetic beads separation
- CD34purity > 90%

※frozen CBs are checked for infectious agents (HBs-Ag, HBc-Ab, HCV-Ab, HIV-I/II-Ab, HTLV-1-Ab, Syphilis (TPHA,RIA)), and proved to be sterile.

INFORMATION CENTER UPGRADING PROGRAM Information

Core Facility : Genetic Resources Center, National Institute of Genetics
Principal Investigator : Yukiko Yamazaki FAX : +81-55-981-6886
Contact site : yyamazak@nig.ac.jp
URL : http://www.nbrp.jp/

Purpose

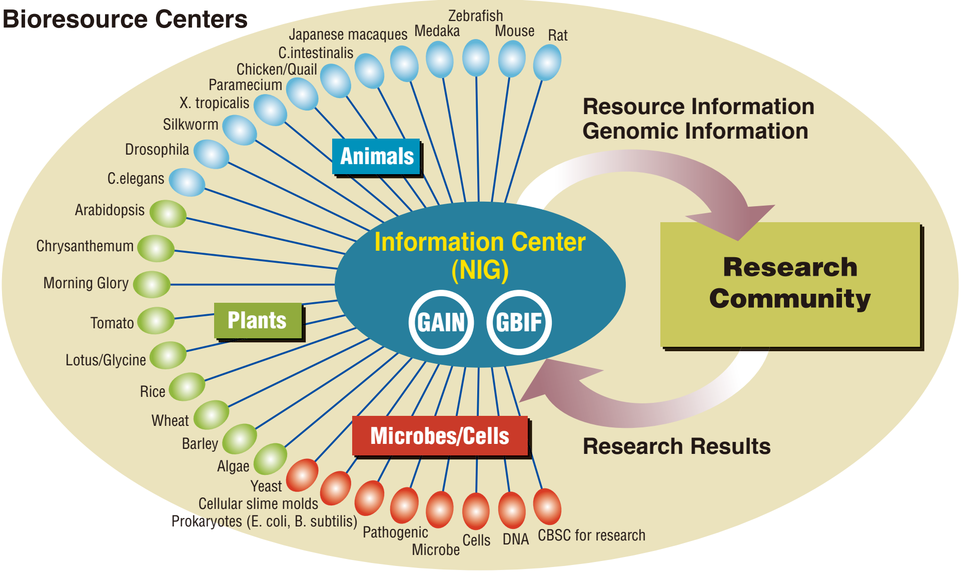
The purpose of the Information Center is to broadly support life science research by promoting the effective use of NBRP core facilities' bioresources through consolidation and provision of information on the bioresource collections' locations, scientific knowledge of the resources, genomic information, and other essential information.There are three key focuses for the implementation of this program: (1) comprehensive collection of information, (2) development of a database for each resource, and (3) effective distribution of information to users.

The Information Center has already made publicly available the information on the bioresources of all core facilities that participated in Phase 1 (2002-2006) and Phase 2 (2007-2011) of the NBRP. In Phase 3 (2012-2016), the Information Center is continuing to provide this information, and is endeavoring to support a positive growth cycle between the resources and research by serving as a nexus that links the user community with the resource provider.

Half a million users access the database of 6.5 million bioresources in 2016, and some 18,800 papers have been published on the results of studies that used the NBRP's collection of bioresources and information.

This program also covers the Information Center's activities pertaining to the Great Ape Information Network (GAIN) and its activities as Japan's node for the Global Biodiversity Information Facility (GBIF).

NBRP Information Center Upgrading Program Implementation System



Resource Research Circulation

GENOME INFORMATION UPGRADING PROGRAM

Genome resequencing of Japanese wild mouse-derived MSM/Ms strain

Core Facility : Genetic Strains Research Center, National Institute of Genetics
Principal Investigator : Toyoyuki Takada FAX : +81-55-981-6817
Contact site : ttakada@nig.ac.jp
URL : http://molossinus.lab.nig.ac.jp/msmdb/index.jsp

Overview

The laboratory mouse is one of the most important bioresources for studying principles underlying higher-order biological phenomena and etiologies of human diseases. National Institute of Genetics (NIG) has established a series of mouse experimental strains named "Mishima Battery" since 1970's. It consists of ten inbred strains originated from three or four subspecies of *Mus musculus*, which are widely distributed in the world. These strains have very remote genetic status from the commonly used laboratory mouse strains such as C57BL/6J (B6), and show very unique phenotypes. The mouse strains in "Mishima Battery" are now distributed to the science research community via RIKEN BioResource center, and contribute to broad range of life science. NIG also established B6-ChrN^{MSM} consomic strains, in which every chromosome of B6 is substituted by corresponding chromosome of Japanese wild mouse-derived MSM/Ms (MSM) strain, which is a member of "Mishima Battery".

For genome analysis of MSM, we previously performed a resequencing project of MSM, which was supported by "NBRP genome information upgrading program" and KAKENHI on Priority Areas "Comparative Genomics". Our study revealed more than ten millions of nucleotide substitutions between B6 reference genome and the MSM genome. In this program, to elucidate more in detail of structure and copy number variation, further resequencing by single molecule and real-time sequencing system is employed for the MSM genome as collaboration with Comparative Genomics Laboratory, and Advanced Genomics Center, NIG, and Joint Support-Center for Data Science Research, ROIS. Upcoming resequenced data will be open to public to increase usability of the MSM genome information for the research community via NIG mouse genome database "NIG_MoG" (http://molossinus.lab.nig.ac.jp/msmdb/index.jsp).

GENOME INFORMATION UPGRADING PROGRAM

Whole genome resequencing of the representative rat strains and development of a SNP typing kit

Core Facility : Medical Institute of Bioregulation, Kyushu University
Principal Investigator : Mikita Suyama FAX : +81-92-642-6692
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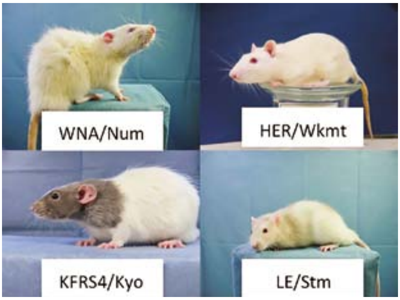
URL : http://www.bioreg.kyushu-u.ac.jp/labo/bioinfo/

Overview

Rats have been used as animal models of human diseases such as cancer and hypertension. NBRP-Rat is the largest repository for the rat strains, and currently about 700 strains are registered in the repository. Among those strains, there are many rat models of immune-mediated disorders and cancers.

This project focuses on whole genome resequencing of the representative rat strains, which include 12 strains of the representative inbred rats, one strain of the inbred rats originated from the wild populations, and seven strains of disease models derived from selective breeding. Based on the SNP information obtained by the whole genome resequencing, we will also develop a SNP typing kit that can be used for linkage analyses to detect responsible loci of diseases and for quality control of registered strains by detecting genetic contamination. This project is conducted in the collaboration with The Institute of Laboratory Animals, Graduate School of Medicine, Kyoto University, which is the NBRP Center for rat strains, and Kazusa DNA Research Institute. The genomic data obtained by this project, together with the development of a SNP typing kit based on the genomic data, will be of help to the researchers in the field of rat genetics.



GENOME INFORMATION UPGRADING PROGRAM

Genome Re-sequencing of Diverse Strains of *Bombyx mori* and *B. mandarina* (2)

Core Facility : The University of Tokyo
Principal Investigator : Toru Shimada Fax : +81-3-5841-8011
Contact site : toru@ss.ab.a.u-tokyo.ac.jp
URL : http://silkbase.ab.a.u-tokyo.ac.jp/

Overview

In the Silkworm NBRP at Kyushu University, about 750 strains are maintained, and distributed to many researchers. Among them, p50T was utilized for whole genome sequencing, and its accurate genome assembly was published in 2008. Nevertheless, the genomes of other strains have not yet been sequenced. In the same program, last year (2015), we selected 18 *Bombyx mori* strains and 2 *B. mandarina* ones, analyzed their genomes by an Illumina sequencer, and released the resultant sequences in DDBJ and GenBank. This year (2016), we will perform highly accurate sequencing of the genome of a Japanese, inbred strain (Sakado) of *B. mandarina* by using a single molecule realtime sequencer (PacBio). *B. mandarina* is the ancestral species of *B. mori*, and exhibits "wild" phenotypes including mimicry in larvae and flight ability in adults, which have been lost in *B. mori* during domestication. On the other hand, *B. mandarina* produces smaller cocoons than *B. mori*. Although *B. mori* and *B. mandarina* can be interspecifically hybridized and produce their fertile progenies, their karyotypes are different; n = 28 in *B. mori* and n = 27 in *B. mandarina*. Therefore, *B. mandarina* is a valuable resource for comparative genomics in both phenotypic and cytological points of view, and its genomic information will be useful for researchers.

Bombyx mandarina **(Wild species of the domesticated silkworm, *B. mori*)**

GENOME INFORMATION UPGRADING PROGRAM

Genome sequencing project of heterocystous cyanobacteria in the NIES collection

Core Facility : Toyohashi University of Technology
Principal Investigator : Yuu Hirose Fax : +81-532-44-6929
Contact site : hirose@ens.tut.ac.jp
URL : http://www.tut.ac.jp/university/faculty/ens/703.html

Overview

Cyanobacteria are photosynthetic bacteria that perform oxygen-evolving photosynthesis. In 1990s, the complete genome sequence of *Synechocystis* sp. PCC 6803 was reported as the first genome of photosynthetic organism (Kaneko et al 1996 *Plant Cell Physiol.*). Since then, genomes of several cyanobacterial strains have been sequenced, and researchers utilize them as model organisms. However, the spread of the next-generation sequencers in 2010s enables us to study much more diverse cyanobacterial strains deposited in the culture collections (Shih et al 2013 *Proc. Natl. Acad. Sci.*).

We focused on the filamentous cyanobacteria that are capable of forming heterocysts. Heterocysts are differentiated cells to fix nitrogen during nitrogen starvation. The genome sizes of heterocystous cyanobacteria are 6-12 Mbp, which are much larger than other non-heterocystous cyanobacteria. In this program, we selected 20 strains of heterocystous cyanobacteria of the National Institute of Environmental Science (NIES) culture collection in Japan (Fig. 1). Their genomes are sequenced and assembled in Toyohashi University of Technology. Annotation and data release are performed in National Institute of Genetics. This program will facilitate both basic and applied studies using cyanobacteria.

Fig1, The organization of this program

FUNDAMENTAL TECHNOLOGY UPGRADING PROGRAM

Development of a new cryopreservation method for Drosophila stocks

Core Facility : Kyoto Institute of Technology
Principal Investigator : Toshiyuki Takano FAX : +81-75-861-0881
Contact site : fruitfly@kit.ac.jp
URL : http://www.dgrc.kit.ac.jp/japanese/

Overview

World-wide Stock Centers such as Kyoto Stock Center, NIG Fly, and Bloomington Drosophila Stock Center house, in total, over 150,000 Drosophila stocks, all of which are maintained by manual labor, that is, transfer of adult flies to a new vial every 2 to 3 weeks. This allows us prompt shipment of flies upon request, but also imposes the upper limit of our capacity. Development of useful and efficient methods for cryopreservation would therefore be of great benefit. Although there are several reports of successful cryopreservation in other non-model insects, we have not yet established a reliable and scalable method for cryopreservation in Drosophila. Because of its large size and a great amount of yolk, it is not easy to cryopreserve embryos; large ovary and testis are also not suitable. In this program, we, in collaboration with University of Tsukuba, aim to develop a new cryopreservation method for Drosophila that can be applied to a wide variety of stocks in the Stock Centers. What's more, this may open new avenues for genome editing applications.

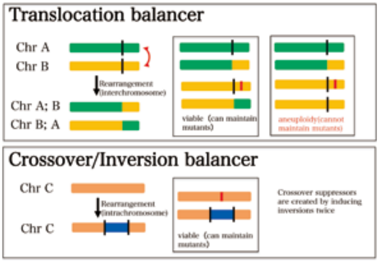
FUNDAMENTAL TECHNOLOGY UPGRADING PROGRAM

Construction of High-Performance balancers for *C. elegans*

Core Facility : Tokyo Women's Medical University School of Medicine
Principal Investigator : Shohei Mitani FAX : +81-3-5269-7414
Contact site : mitani.shohei@twmu.ac.jp
URL : http://shigen.lab.nig.ac.jp/c.elegans/

Overview

The nematode *Caenorhabditis elegans* is the most simple model animal. Because the life cycle is 3 days at 20˚C, the animals are very useful for genetic analyses. We have been distributing deletion mutants from the first term of NBRP and many papers have been published. Roughly 20% of the mutants show lethal or sterile phenotypes and they may be diluted by the wild type animals and eventually lost during short culture periods. To maintain such mutants, we are using balancers. However, there are chromosomal regions where balancers are missing, and many of the popular balancers are translocation type and suffer from lethal phenotype because of aneuploidy. As shown in the figure, balancers of crossover/inversion type, which have rearrangement within the same chromosome are very useful. We are generating stable crossover-type balancers by inducing inversions twice in overlapped regions of a chromosome,using the genome editing technology. We label those balancers by integrating fluorescent protein reporter transgenes for convenience of researchers. We will generate such ideal balancers for whole *C. elegans* chromosomes.



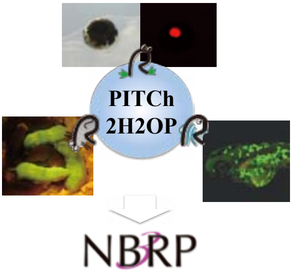
FUNDAMENTAL TECHNOLOGY UPGRADING PROGRAM

Development of easy protocols for efficient gene knock-in using genome editing technology

Core Facility : Graduate School of Science, Hiroshima University
Principal Investigator : Takashi Yamamoto FAX : +81-82-424-7498
Contact site : tybig@hiroshima-u.ac.jp
URL : http://www.mls.sci.hiroshima-u.ac.jp/smg/index.html

Overview

Gene knock-out strategies have become commonplace in various model organisms since the introduction of the CRISPR-Cas9 system. Gene knock-in strategies, which can precisely insert a desired modification into any target site in a genome, are extremely powerful for creating specific mutant lines; however, current knock-in protocols are unsatisfactory because they are inefficient and highly laborious. In this subject, Hiroshima university in collaboration with Osaka university and Yamanashi university aims to develop easy protocols for efficient gene knock-in, which can be applied to various model organisms using two techniques: Precise Integration into Target Chromosome (PITCh), based on Microhomology-Mediated End Joining (MMEJ) repair, and a Two-Hit Two-Oligo with Plasmid (2H2OP) system using single-stranded oligonucleotides as ‘paste’. We optimize these two techniques to increase knock-in efficiency and to reduce mosaicism and off-target effects in three model animals, rat (*Rattus norvegicus*), frog (*Xenopus tropicalis*), and zebrafish (*Danio rerio*). Finally, for National BioResource Project (NBRP) users and in collaboration with NBRP Core Facility Upgrading Programs, we provide an optimized gene knock-in protocol and valuable information regarding genome editing.



FUNDAMENTAL TECHNOLOGY UPGRADING PROGRAM

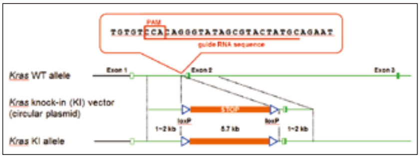
Fundamental technology development of genome editing for the establishment of intractable disease models

Core Facility : Experimental Animal Division, RIKEN BioResource Center
Principal Investigator : Atsushi Yoshiki FAX : +81-29-836-9010
Contact site : animal@brc.riken.jp
URL : http://mus.brc.riken.jp/en/

Overview

Since sequence analysis of genetic factors of human diseases in patients is rapidly advancing in recent years, it is required immediately to analyze the effects of causative mutations for intractable diseases on the pathogenesis in the whole body. In this study, we plan to develop the efficient method to generate knock-in mice by genome editing and create useful mouse models to investigate refractory cancers, which will contribute to advanced quality control of genetically-modified mice created by genome editing knock-in technology. Our group will develop modified Cas9 functional molecule with optimization for homology-directed repair and establish a method for genome-edited knock-in mouse generation by pronuclear microinjection. In addition, we will generate following four strains by using the above genome editing knock-in technology in order to establish useful and basic genetic tools to develop mouse models of refractory cancers such as pancreatic and lung cancers: conditional strain carrying point mutations of *Kras* which are highly lethal in heterozygotes, conditional *p53* knockout strain which is lethal in homozygotes, pancreatic-tissue specific inducible Cre strain and Cre-dependent high-intensity luciferase reporter strain. All the strains generated in this study will be deposited at the NBRP core facility of the mouse and distributed to the scientific community.

Mice are widely used in development of human disease models



Schema of genetic modification of Kras gene by genome editing knock-in

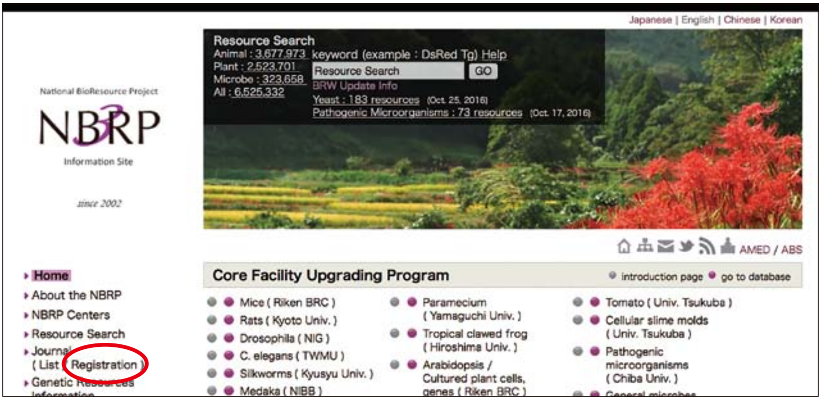
NBRP needs feedback from the users.

Accumulation of research outcome using bioresources can further enhance the value of the bioresouces. NBRP is collecting such research outcomes, and integrating them into the NBRP database. Therefore, we would like to request the bioresource users 1) to describe “the name of the bioresource and its supplier” in the research papers and 2) to send the paper information to the NBRP Core Facility, upon publication of research outcome using the NBRP resources.

Please visit to the “Research Paper Online Registration Site” (see below) for easy feedback of such information.

Please click “Journal (List / Registration)”, on the top page of

http://www.nbrp.jp/



Resource Search

Animal : 3,677,973

Plant : 2,523,701

Microbe : 323,658

All : 6,525,332

keyword (example : DsRed Tg)

BRW Update Info

Yeast : 183 resources (Oct. 25, 2016)

Pathogenic Microorganisms : 73 resources (Oct. 17, 2016)

Core Facility Upgrading Program

- Mice (Riken BRC)
- Rats (Kyoto Univ.)
- Drosophila (NIG)
- C. elegans (TWMU)
- Silkworms (Kyusyu Univ.)
- Medaka (NIBB)
- Paramecium (Yamaguchi Univ.)
- Tropical clawed frog (Hiroshima Univ.)
- Arabidopsis / Cultured plant cells, genes (Riken BRC)
- Tomato (Univ. Tsukuba)
- Cellular slime molds (Univ. Tsukuba)
- Pathogenic microorganisms (Chiba Univ.)

How to order and about handling/shipping costs

How to order:

Please go to the NBRP homepage (http://www.nbrp.jp), and click on the resource name you wish to get, which is listed on the left side of the homepage. Then, follow the instructions to proceed.

About handling and shipping costs:

The expenses for handling and shipping will be charged to the recipients.

Education activities and consultation system on the Nagoya Protocol

At the 10th Conference of Parties (COP10) to the Convention on Biological Diversity in October 2010 in Nagoya, Japan, the Nagoya Protocol on Access to Genetic Resources and the Fair and Equitable Sharing of Benefits Arising from their Utilization (hereinafter referred to as "Nagoya Protocol") to the Convention on Biological Diversity was adopted. It entered into force on 12 October 2014.

The ABS system and the Nagoya Protocol also apply to non-commercial academic research. Academic and public research institutions seek access to and use of genetic resources and traditional knowledge related to biological resources to increase our understanding of nature. Here are our major activities based on the actual corresponding to the Nagoya Protocol.

1. Conduct education and public information activities (holding of seminars, workshops, symposiums etc.)
2. Information distribution services (making and distributing leaflets, posters etc.)
3. Build support system (establishment of the support office)


Mutsuaki Suzuki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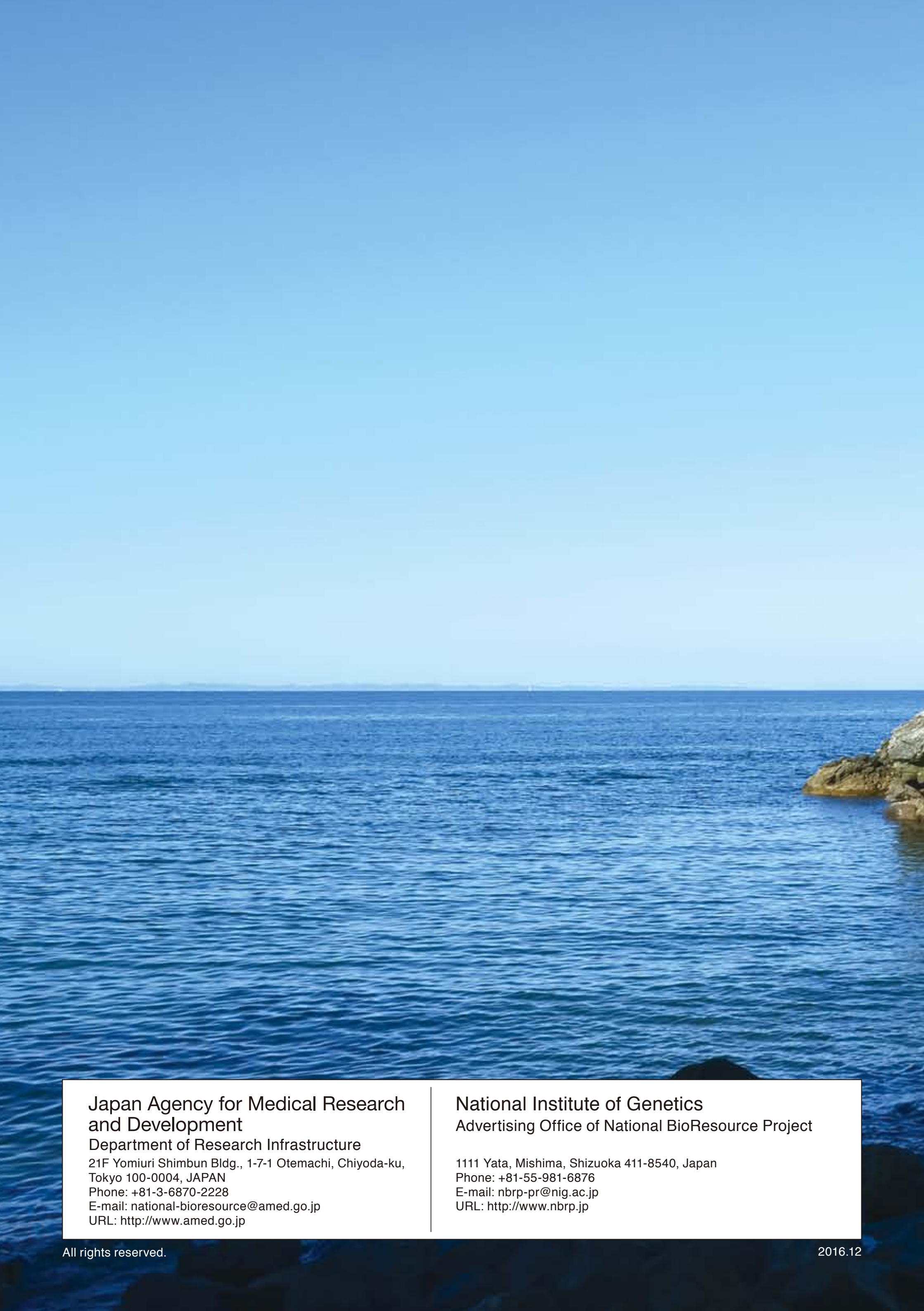
Website for the ABS Task Force Team for Academia

植物・動物・微生物を海外で入手し研究に利用する時には、生物多様性条約への対応が必要です



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