

Departmental Overview

Responsibility and duties of collecting and storing biosamples at ABCC and RERF had been mostly assigned to individual research departments until recent years. To preserve precious biosamples such as blood (serum, plasma, and cells), urine, pathological specimens, blood smears, and teeth, in good condition, and to promote research use of those biosamples, it was considered essential to centralize their management and to generate a comprehensive biosample database. In 2013, the Biosample Research Center was established to undertake these tasks. With the goal of clarifying late effects of ionizing radiation on medical conditions of A-bomb survivors and their children, and of elucidating their molecular mechanisms, the Center is arranging appropriate preservation to ensure effective utilization of those invaluable materials, donated by A-bomb survivors, and their spouses and children. To achieve these objectives, biosamples and associated information previously collected by individual departments have been transferred to the Center, whereas most of the biosamples currently collected by RERF have been processed and stored by the Center with new standard operating procedures (SOPs) since 2015.

Because freezer rooms in Hiroshima Lab had been filled beyond the capacity with 51 upright deep freezers full of samples, RERF installed a robotic deep-freezer biorepository system (BioStore II) in 2015 to accommodate about 592,000 archival sample tubes compatible with the system and to efficiently manage future blood and urine samples. The relocation of the archival biosamples from conventional freezers to the BioStore II was initiated in 2017 and finalized in 2021.

Quality assurance and quality control (QA/QC) of both aged and new biosamples is essential to ensure accuracy of data obtained by any analytical methods. In 2021, the Center established methods for QA/QC of blood plasma by proteomic analysis using a mass spectrometer.

Development of the Biosample Research Center and the Research Resource Center (RRC) is one of the top priorities of RERF to facilitate internal and external collaborative research. To make most of the precious biosamples, it is planned that the RRC will link the inventory and quality information of biosamples with epidemiological and clinical information to construct an integrated annotated biosample database of RERF, with which internal and external researchers can search for and find biosamples necessary for their research. Therefore, the Center needs to generate and constantly update a comprehensive biosample database and continue providing it to the RRC. In 2020, to record and manage all workflows as well as the inventory and quality information of biosamples, and to generate a biosample database, the Center designed and implemented a laboratory information management system (LIMS). In 2021, to prepare for sample provision to both internal and outside researchers, the Center finalized detailed procedures and forms for sample provision that were brought into effect, and also customized and upgraded the LIMS for the management of workflows and information related to biosample provision in accordance with the procedures and forms.

As a future plan, the Center will contribute to the development of a Sample Analytical Unit (SAU) with in-house capabilities in omics and image analysis to conduct QA/QC of biosamples, serve as a resource to drive both internal and external collaborative research, and minimize consumption of precious biosamples.

FY2021 Biosample Research Center Achievements

The Center finalized detailed procedures for sample provision and sample request forms, and

had them approved by the Committee on Biosamples and the Executive Committee to go into effect. In accordance with the procedures and forms, the Center customized and upgraded the LIMS (laboratory information management system) for the management of workflows and information related to biosample provision for research use.

Storage and Inventory Management of Biosamples

- Since its inauguration in 2013, the Center has prepared and been updating standard operating procedures (SOPs) for receipt, processing, and storage of biosamples according to the guidelines by ISBER (International Society for Biological and Environmental Repositories) and OECD (Organisation for Economic Co-operation and Development).
- During the past year (Dec. 2020 – Nov. 2021), the Center newly processed and stored blood and urine samples provided by 596 AHS subjects (275 subjects at Hiroshima Lab, hereafter referred to as "H," and 321 at Nagasaki Lab, hereafter referred to as "N"), or by 1,637 FOCS subjects (H: 1,007, N: 630). A total of 42,470 tubes of blood samples were processed and stored; 24,159 tubes were by Hiroshima Center, of which 7,588 were transported to Nagasaki for backup storage, and 18,311 tubes were processed by Nagasaki Center, of which 5,726 were stored in Hiroshima. The Center also newly processed and stored 12,544 tubes of urine samples (H: 7,200, N: 5,344).
- Since 2015, the Center has processed and stored a total of 428,000 tubes of blood samples (H: 277,000, N: 151,000) and 93,000 tubes of urine samples (H: 61,000, N: 32,000), provided by 3,595 AHS subjects (H: 2,207, N: 1,388) and 9,850 FOCS subjects (H: 6,779, N: 3,071) as of Nov. 2021.
- As for archival samples, since 2014 the Center has inventoried and acquired archival blood and urine samples of AHS and FOCS subjects, which had been collected and preserved by the Departments of Clinical Studies and Molecular Biosciences (MBS). Currently, the Center holds a total of about 1,309,000 tubes of archival blood samples (H: 874,000, N: 435,000) and 157,000 tubes of archival urine samples (H: 100,000, N: 57,000), which had been provided by 16,803 AHS subjects (H: 11,263, N: 5,540) since 1969, and by 12,689 FOCS subjects (H: 8,489, N: 4,200) since 2002.
- Since 2018, the Center has been extracting DNA from archival blood clot samples provided by AHS subjects from 2003 to 2013 by using an automated DNA extractor named MagCore (RBC Bioscience) and has stores a total of 6,254 DNA samples from 4,232 AHS subjects (H: 3,800, N: 432) as of Nov. 2021.
- Blood cells donated by Trio families have been preserved by MBS in liquid nitrogen tanks and deep freezers since 1985 for genetic/genomic studies on trans-generational effects of parental radiation exposure. In 2020, the Center and MBS jointly completed inventory of a total of 58,980 tubes of blood samples (fresh frozen mononuclear cells 11,109, EBV-transformed lymphocytes 41,426, and granulocytes 6,445), which were provided by 4,140 individuals including 1,004 Trio families comprising A-bomb survivors and their spouses and 1,653 children. These samples have been transferred to the Center for centralized management and efficient research use.
- As of Nov. 2021, the Center holds a grand total of about 2,053,000 tubes of biosamples comprising about 1,796,000 tubes of blood samples (H: 1,169,000, N: 627,000), about 251,000 tubes of urine samples (H: 161,000, N: 89,000), and 6,254 tubes of DNA samples (H: 6,254, N: 0) including both archival samples and those processed and stored by the Center, which have been provided by 16,813 AHS subjects (H: 11,266, N: 5,547), by

12,786 FOCS subjects (H: 8,522, N: 4,264), and by 4,140 Trio study subjects.

- Since 2017, the Center of Hiroshima Lab has been relocating inventoried archival blood and urine samples from conventional upright freezers to the robotic deep-freezer biorepository system (BioStore II). During 2021, the Center transferred 12,162 more tubes and thus almost finalized the relocation of all the 591,726 tubes compatible with the robotic system. The archival sample tubes now occupy 56% of the storage capacity of the BioStore II.
- In 2020, the Center designed, optimized, and implemented a commercial LIMS, LabVantage[®], to manage biosample workflows such as receipt, processing, storage, and transport of biosamples, to manage inventory and quality information of biosamples, to manage reagent and consumable inventories, and to generate a comprehensive biosample database. In 2021, the Center further customized LIMS for the management of workflows and information related to biosample provision for research use (see below).
- Over 500,000 formalin-fixed paraffin-embedded (FFPE) blocks and over 900,000 glass slides of pathological tissue specimens from about 13,000 cases of autopsies and 130,000 cases of surgical procedures are preserved in the Pathology Laboratories of the Department of Epidemiology both at Hiroshima and Nagasaki Labs. The Pathology Laboratories have been conducting the inventory of those archival samples to allow for efficient research use. The FFPE blocks are being indexed, ordered, and packaged, and the numbers of available samples and organs of origin are being recorded in a database. The glass slides are being identified, counted, linked with FFPE blocks, and recorded in the database. Since 2020, the Center has been assisting the Pathology Lab at Hiroshima Lab with inventorying FFPE blocks to near completion and is now assisting the inventory of glass slides as well.

Preparation for Biosample Provision

- The Center finalized detailed procedures for sample provision and sample request forms and had them approved by the Committee on Biosamples and the Executive Committee to go into effect. In accordance with the procedures and forms, the Center also customized and upgraded the LIMS for the management of workflows and information related to biosample provision for research use.

Quality Assurance and Quality Control of Biosamples

- The Center developed methods for quality assurance and quality control (QA/QC) of blood plasma by a liquid chromatography tandem mass spectrometer (LC-MS/MS), SCIEX TripleTOF 6600+, procured in 2020. We identified several plasma proteins that may serve as useful quality markers for retrospective estimation of a pre-centrifugal delay (time interval between blood drawing and centrifugation in the process of plasma sample preparation) by evaluating effects of the pre-centrifugal delay on global quantification of trypsin-digested peptides derived from plasma proteins by using LC-MS/MS.
- The Center has developed Business Continuity Plans as a trouble shooting system to be well prepared against electric blackout, liquid nitrogen shortage, network failure, and the likes, caused by disasters including storm, earthquake, landslide, fire accident, and national or global pandemic that can cause unexpected shortage of manpower and supplies.