

BIOSAMPLE RESEARCH CENTER

Responsibility and duties of collecting and storing biosamples at ABCC/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, and blood smears, 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 aims to reveal late effects of ionizing radiation on medical conditions among A-bomb survivors and their children, and to elucidate 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, while 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 deep freezers packed with blood and urine samples, a robotic deep-freezer biorepository system (BioStore II) was installed in 2015 to accommodate about 592,000 archival sample tubes compatible with the system and to efficiently manage future biosamples. The transfer of the archival biosamples from conventional freezers to the BioStore II was initiated in 2017, and completed in 2021.

Quality assessment of both aged and new biosamples is essential to ensure accuracy of data obtained by any analytical methods. The Center has established methods for quality assessment of blood plasma by proteomic analysis with a mass spectrometer.

The development of the Biosample Research Center and the establishment of the Research Resource Center (RRC) are the top priorities of RERF to promote internal and external collaborative research. To make the 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 generate an integrated research database of RERF, with which internal and outside researchers can easily search for and find biosamples available to their research. Therefore, the Center needs to generate and constantly update a comprehensive biosample database, and keep 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 the 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 prepared detailed procedures and forms for sample provision, and has also customized and improved 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 a Sample Analytical Unit (SAU) that will be equipped with in-house capabilities in omics and image analysis to conduct sample quality assessment of biosamples, and serve as a resource to drive both internal and external collaborative research, and to minimize consumption of precious biosamples.

FY2022 Biosample Research Center Achievements

The Center had finalized detailed procedures for sample provision and sample request forms, and had them go into effect. In accordance with those procedures and forms, the Center has been customizing and improving 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. 2021–Nov. 2022), the Center newly processed and stored blood and urine samples provided by 576 AHS subjects (317 subjects at Hiroshima Lab, hereafter referred to as "H", and 259 at Nagasaki Lab, hereafter referred to as "N"), or by 2,082 FOCS subjects (H: 1,450, N: 632). A total of 53,086 tubes of blood samples were processed and stored; 35,466 tubes were by Hiroshima Center, of which 10,290 were transported to Nagasaki for remote backup storage, and 17,620 tubes were processed by Nagasaki Center, of which 5,203 were transported to Hiroshima. The Center also newly processed and stored 20,945 tubes of urine samples (H: 13,957, N: 6,988).
- Since 2015, the Center has processed and stored a total of 481,000 tubes of blood samples (H:312,000, N: 169,000) and 114,000 tubes of urine samples (H: 75,000, N: 39,000), provided by 3,597 AHS subjects (H: 2,208, N: 1,389) and 9,893 FOCS subjects (H: 6,801, N: 3,092) as of Nov. 2022.
- 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: 881,000, N: 428,000) and 157,000 tubes of archival urine samples (H: 100,000, N: 57,000), which had been provided by 16,802 AHS subjects (H: 11,263, N: 5,539) since 1969, and by 12,689 FOCS subjects (H: 8,489, N: 4,200) since 2002.
- Since 2018, the Center has extracted DNA from archival blood clot samples provided by AHS subjects from 2003 to 2013 by using an automated DNA extractor, MagCore (RBC Bioscience), and stored a total of 8,149 DNA samples from 6,127 AHS subjects (H: 3,800, N: 2,327) as of Nov. 2022.
- Blood cells donated by Trio families have been preserved by MBS in liquid nitrogen tanks and deep freezers since 1985 for genetic studies on trans-generational effects of parental radiation exposure. In 2020, the Center and MBS jointly inventoried a total of 58,980 tubes of blood samples (11,109 tubes of fresh frozen mononuclear cells, 41,426 of EBV-transformed lymphocytes, and 6,445 of granulocytes), which were provided by 4,140 individuals including 1,004 complete sets of trios comprising AHS subjects, their spouses, and 1,653 children who are F1 Study subjects. These samples were transferred to the Center for centralized management and efficient research use.

- As of Nov. 2022, the Center holds a grand total of about 2,121,000 tubes of biosamples comprising about 1,849,000 tubes of blood samples (stored in H: 1,199,000, N: 650,000), about 272,000 tubes of urine samples (stored in H: 175,000, N: 96,000), and 8,149 tubes of DNA samples (stored in H) including both archival samples and those processed and stored by the Center, which have been provided by 16,802 AHS subjects (provided at H: 11,262, N: 5,540), by 12,740 FOCS subjects (provided at H: 8,501, N: 4,239), and by 4,140 Trio study subjects (provided at H: 2,224, N: 1,916).
- From 2017 to 2021, the Center of Hiroshima Lab transferred about 592,000 tubes of inventoried archival blood and urine samples from conventional upright freezers to the robotic deep-freezer biorepository system (BioStore II) in Hiroshima. The archival sample tubes now occupy 55% of the storage capacity of the BioStore II. In addition, 311,000 tubes of samples processed and stored by the Center since 2015 are also housed in BioStore II, occupying 8% of its capacity, leaving 36% unoccupied.
- In 2020, the Center designed, optimized, and implemented a LIMS, LabVantage[®], to manage biosample workflows such as receipt, processing, storage, and transportation of biosamples, to manage inventory and quality information of biosamples, to manage inventories of reagents and consumables, and to create a comprehensive biosample database. Since 2021, the Center has been further customizing the 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 effective research use. The FFPE blocks have been 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. Since 2020, the Center has been assisting the pathology laboratory at Hiroshima Lab with inventory of FFPE blocks to completion, and is now assisting inventory of glass slides.

Biosample Provision

- In FY2021, the Center finalized detailed procedures for sample provision and sample request forms, and had them go into effect. Since then, in accordance with the procedures and forms, the Center has been customizing and improving the LIMS for the management of workflows and information related to biosample provision for research use.
- The Center provided blood samples of AHS participants for a Research Proposal titled "Preliminary study to determine the applicability for GWAS of DNA extractable blood smears and blood-infiltrated paper discs preserved in the past " (by T. Hayashi, et al.) with the procedures and forms, formalized as described above. Those samples are blood-infiltrated paper discs preserved at -80°C for about 20 years and provided by 12 AHS participants who gave re-consent to the use of those samples, and will be tested for their applicability to array-based SNP typing.

Quality Assessment of Biosamples

- Since 2020, the Center has been developing methods for quality assessment of blood plasma and serum by a quadrupole time-of-flight mass spectrometer (QTOF-MS), SCIEX TripleTOF 6600+. Initially by global non-targeted quantification of enzymatically digested peptides of plasma proteins using the QTOF-MS, we identified several candidate peptides that may serve as quality markers for past events of freezing and thawing, and then tested the validity of those candidate peptides as useful quality markers by targeted quantification by high-resolution multiple reaction monitoring (MRM-HR) with the QTOF-MS.
- In 2020, the Center developed Business Continuity Plans (BCP) as damage control plans in order for us 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, and has been continuously updating the BCP.