



MEMORANDUM

DATE: October 3, 2025
TO: Network Clinical Trial Units
FROM: IQA Cryopreservation Proficiency Testing Program
SUBJECT: Reminder Notice for the IQA Cryopreservation Proficiency Testing Program

Sites/Labs Participation

The use of cryopreserved Peripheral Blood Mononuclear Cells (PBMC) for immunologic assays has increased in recent years with the expansion and diversification of sites participating in DAIDS-supported network studies. The utility of cryopreserved PBMC for efficient immunological assays is predicated on their viability and functional ability in vitro.

The leaderships of the Advancing Clinical Therapeutics Globally for HIV/AIDS and Other Infections (ACTG) and the International Maternal Pediatric Adolescent AIDS Clinical Trials Group (IMPAACT) require laboratories that cryopreserve PBMCs for network protocols to participate in the Immunology Quality Assessment (IQA)-administered quarterly proficiency testing program to determine their ability to reliably freeze viable PBMCs.

If your site will not be able to submit samples this iteration of the program, please send a message to Raul Louzao (raul.louzao@duke.edu) or Carmela Archual (carmela.archual@duke.edu) no later than **December 12, 2025** with an explanation of your circumstances. In your message, please include the specific ACTG/IMPAACT site name and LDMS Lab number unable to participate. The network(s) will determine whether or not your lab will be placed “on hold” for a maximum of two rounds of IQA testing. While your lab is “on hold” it is not allowed to cryopreserve PBMC specimens for ACTG or IMPAACT protocols.

Participating Labs:

We will start receiving samples the week of **December 1, 2025**.
Samples will be considered **LATE** after **December 12, 2025**.

Prepare Samples

1. Identify one (1) blood donor for each participating processor and obtain their consent:
 - a. Submission of HIV-positive PBMC samples is no longer required; laboratories may now submit HIV-negative PBMC samples for scoring. In other words, a site may now choose to submit cells from either HIV+ or HIV- donors but must indicate the HIV serostatus of the donor. Labs do not need to provide documentation of the HIV- serostatus.
 - b. A maximum of 2 PBMC processors can submit each quarter. Laboratories with only 1 PBMC processor will only submit 1 donor, while laboratories with more than 2 PBMC processors are expected to maintain a regular rotation of up to 4 processors, with 2 sets of 2 technicians submitting samples in alternating quarters.
2. Arrange to obtain blood from the volunteer(s) as early as the week of **November 3, 2025** (Samples must be stored frozen for at a minimum of 1 week, and a maximum of 5 weeks before shipping to the IQA).



3. From each donor, obtain at least 20mL of blood by standard venipuncture technique. ACD, EDTA or Heparin may be used as the anticoagulant.
4. Isolate PBMC's from each donor according to the Cross-Network PBMC Processing SOP following the protocol in which you currently process PBMCs; this can be downloaded at <https://www.hanc.info/resources/sops-guidelines-resources/laboratory/cross-network-pbmc-processing-sop.html>.
 - a. Determine the cell yield and viability by trypan blue dye exclusion.
 - b. Both % viability and viable cell recovery will be assessed by the IQA after thawing, so an accurate count of total viable cells in each vial is important.
5. You should have four aliquots for each donor. Each aliquot should contain a viable cell concentration of **~5 x 10⁶ cells per cryovial**, but concentrations as low as 3 x 10⁶ cells per cryovial are accepted.

Note:

- If the total viable cells are less than 20 x 10⁶ cells but greater than 12 x 10⁶ cells, divide the PBMC into four equal aliquots and be sure the LDMS label reflects the actual number of cells per vial.
- If the total viable cells are less than 12 x 10⁶ cells, divide the PBMCs into two equal aliquots and ship to the IQA.
- **DO NOT SHIP VIALS CONTAINING LESS THAN 3 x 10⁶ cells per vial OR VIALS CONTAINING MORE THAN 10 x 10⁶ cells per vial.**

6. **The freezing process and cell storage should be the same as the freezing process and cell storage your lab uses for study participant samples.**

Note: If you cryopreserve PBMC for both ACTG and IMPAACT, you might choose to use one of these options:

- Use the freezing and storage methods you would normally use for the network for which you process the largest number of samples
- Alternate between ACTG and IMPAACT methods from round to round
- Within each round of testing, use ACTG methods for the samples from one donor and IMPAACT methods for the samples from the other donor

7. **Ship two aliquots from each donor to the IQA** and store two back-up aliquots from each donor as your lab would normally store study participant samples. **Please do not send additional aliquots.** The back-up aliquots might be needed for future testing or in-lab investigation.

Additional information can be found at <https://dhvi.duke.edu/programs-and-centers/immunology-virology-quality-assessment-center/research-programs/immunology-5>.



LDMS

It is **MANDATORY** that all IQA Cryopreservation and Viability samples be logged in, resulted and shipped through LDMS. Please follow the steps outlined below to ensure that the data has been entered properly. For additional information, please reference the LDMS User Manual sections listed for each step. If you have any questions regarding the LDMS program, please contact LDMS user support at 716-834-0900 ext. 7311 or email ldmshelp@fstrf.org.

Step 1: Log the samples into Specimen Management.

Samples must be logged into the Specimen Management module as group IQA with the PID or donor identifier entered into the ID1 field. For each aliquot, enter the cell count for the volume with CEL as the derivative and the volume unit. The IQA Cryopreservation and Viability assay should be ordered in the Test Setup tab of Specimen Management for each aliquot.

Step 2: Generate Labels for IQA Specimens.

Choose the IQA Barcode Label or use the Labels module for the IQA specimens that will print out the ID1, LDMS specimen #, cell volume, specimen date and derivative type for each label. Make sure the IQA label shows (IQA group provides a fixed IQA Barcode label):

Specimen ID (donor #)
Global Spec ID (LDMS #)
Specimen Date (draw date)
PRIM/ADD/DER (cell)
Cell Volume/ml

Step 3: Complete the IQA Cryopreservation and Viability Data Entry Screen

Go into the Specimen Management module and locate the specimen. From the **Edit** button to the right of the specimen, click the down arrow, and then click **Cryopreservation** (this option is only available for specimens with the derivative type CEL). Complete the information in the **HIV Status**, **Primary Specimen Details**, **Aliquot Details**, and **Technician Details** sections. In the Processing Tech Initials box, enter the processing technician's initials. **Save** the information.

Step 4: Create a Shipping Disk and Manifest Report

In the Shipping module, search for your IQA samples and create a shipping disk to be sent to the IQA **Lab 213**. Alternately, you can create an LDMS Shipping Batch file to be emailed to Lab 213. **These files (from either the shipping disk or shipping Batch file) will need to be imported into Lab 213 and are required to proceed.** Print out and include a copy of the manifest report in your shipment.

IQA Cryo WBS

It is **MANDATORY** that all IQA Cryopreservation PT Program samples be logged into the IQA Cryo WBS. Please refer to the IQA Cryo WBS Users Guide for directions on how to log quarterly samples in the IQA Cryo WBS.



Duke Human Vaccine Institute

Shipping Instructions

Arrange to ship the samples on dry ice, on Monday, Tuesday, or Wednesday during the week of **December 1, 2025 or December 8, 2025.**

Refer to the cross-Network shipping training video online at <https://www.hanc.info/labs/labresources/procedures/Pages/actnShippingDemo.aspx>. Please note that for the IQA Cryopreservation Proficiency Testing Program, you will be shipping a Category B Biological Substance.