

233. LONGITUDINAL PVDOMICS BIOREPOSITORY CORE BLOOD COLLECTION OVERVIEW AND BLOOD DRAWING PROCEDURES

233.1 Purpose and Background

Participants who enroll in the Longitudinal PVDOMICS (L-PVDOMICS) Study will be asked to provide biological samples (blood and urine) for OMICS analysis (Genomics, Proteomics, Metabolomics, and Coagulomics). If participants agree to storage of their biological samples for use in possible future studies, a separate Biorepository informed consent will be signed.

Among those participants who consent for storage of biological specimens, serum, plasma, buffy coat, and urine **samples will be collected at one time only for Level 1 participants**. This document describes the collection of blood specimens. Chapter 234 describes processing and shipping of blood specimens.

Chapter 235 describes collection, processing and shipment of urine specimens.

233.2 Biorepository Contacts

The Biorepository under supervision of Suzy Comhair, PhD, is part of the DCC and is located at the Cleveland Clinic, Cleveland, Ohio. Laura Peterson is the Biorepository manager. Their contact information can be found in the PVDOMICS Address Directory. For general questions, e-mail pvd-biorepos@bio.ri.ccf.org.

233.3 Collection Schedule, Type and Volume of Samples

Venous Blood is collected one time during the L-PVDOMICS study for those participants who enrolled in Level 1 only.

Table 233.1 L-PVDOMICS Blood Samples Volumes

Vacutainer™ blood tubes	L-PVDOMICS
Gold top (SST)	2 x 5 ml
Lavender top (EDTA)	1 x 10 ml
Red top (PAXgene®)	1 x 2.5 ml
TOTAL Blood Collected	22.5 ml

233.4 Personnel Blood Collection Training Requirements

Study personnel certified for blood collection and processing in the parent study is automatically certified for L-PVDOMICS. Persons collecting and processing specimens for L-PVDOMICS who have not been certified for the parent study, will be certified by written exam to ensure samples are uniformly handled prior to analyses. Each PVDOMICS center will designate a study staff member who will be responsible for training the appropriate personnel.

233.5 Supplies for Blood Collection and Shipment

The Biorepository provides sample collection/shipping kits. Ten collection/shipping kits will be shipped to each center at the beginning of the study. Additional kits will be shipped to centers monthly or as needed. The number of the additional kits will be determined by the actual center recruitment. If kits are not received for any reason, or extra supplies are needed, the coordinator needs to contact the Biorepository Core to request additional supplies.

Table 233.2 L-PVDOMICS Blood Collection Kit

Item Number	Description	Quantity
Fisher: 02-683-97 BD: 367986	5 ml Serum Separator Gold top Vacutainer™ blood tube (gel barrier)	2
Fisher: 02-657-32 BD: 366643	10 ml EDTA Plasma Lavender top Vacutainer™ blood tube	1
Fisher: 23-02101 BD: 762165	2.5 ml PAXgene® Red top Vacutainer™ blood tube	1

Standard phlebotomy kits that include items such as: alcohol wipes or swabs, adhesive tape, gloves, tourniquets and 21 gauge needles are not part of the collection kit and must be purchased using local center funds.

The peripheral venous blood collection needs to occur with a 21 gauge needle.

233.6 Preparation for Level 1 PVDOMICS Blood Collection

Blood will be collected one time during the study. Several days prior to each collection, the study coordinator prepares the collection kit for the participant and the data collection forms. Each biorepository kit is identified by a 9 alphanumeric Kit ID: xxLPVDxxx. The first two digits are the participating site number, the next 4 characters note that the kit is for L-PVD, the last 3 digits are uniquely assigned to the Kit by the DCC. The coordinator needs to note the Kit ID selected for a specific participant ID on Form 500 by placing the provided Kit ID label on p.2 on the Form. Once Form 500 is entered in the database, this Kit ID and the barcodes on the vials in it will only be available for this participant ID.

The coordinator needs to check expiration dates on the Vacutainer™ blood tubes included in the kit, and the participant ID and collection number. If the Vacutainer™ blood tubes included the kit have expired or any other issues are discovered, the coordinator needs to contact the Biorepository Core to request additional supplies.

The coordinator contacts the participant the day prior to the collection to remind them to avoid all food and liquids except for small sips of water for pills, for 8-10 hours prior to blood collection. Every effort should be made to schedule the biospecimen collections in the morning to avoid potential diurnal effects of measures.

233.7 Blood Draw Collection Procedure

- o All participants will need to **avoid all food and liquids** except water for 8-10 hours prior to blood collection. Record the number of hours the participant fasted on Form 800.
- o Blood collection needs to occur after vital signs and prior to physical exam.
- o Before beginning the blood collection, participants need to be resting in a chair for 15 min. This time should be in a quiet area with minimal talking and the participant should not be doing questionnaires or other study components.
- o Complete Form 800 to document the blood draw.

Procedure

1. Apply tourniquet.
 - o Wrap the tourniquet around the arm 3 to 4 inches (7.5 to 10.0 cm) above the venipuncture site. Tuck the end of the tourniquet under the last round. If a Velcro tourniquet is used adhere the ends to each other.
2. Identify vein.
 - o Palpate and trace the path of veins several times with the index finger. Unlike veins, arteries pulsate, are more elastic, and have a thick wall. Thrombosed veins lack resilience, feel cord-like, and roll easily – these should be avoided. If superficial veins are not readily apparent, lowering the extremity over the arm of the chair will allow the veins to fill to capacity. Identify the best available vein.

3. Assemble the butterfly-Vacutainer™ set, if butterfly is used. If a straight phlebotomy needle is used, confirm that it is a 21g and then attach it to the Vacutainer™ holder with the Luer adapter.
 - o Attach the Luer adapter to the Vacutainer™ holder.
 - o Attach the Luer end of the butterfly needle (21 gauge needle) set to the Luer adapter.
4. Cleanse the venipuncture site.
 - o Remove alcohol prep from its sterile package.
 - o Cleanse the vein site with the alcohol prep using a circular motion from the center to the periphery.
 - o Allow the area to dry to prevent possible hemolysis or contamination of the specimen and a burning sensation to the participant when the venipuncture is performed.
5. If venipuncture becomes difficult, the vein may need to be touched again with a gloved hand. If this happens, cleanse the site again with alcohol and allow the skin to dry.
6. Perform venipuncture.
 - o Grasp the participant's arm firmly, using your thumb to draw the skin taut. This anchors the vein. The thumb should be 1 or 2 inches (2.5 or 5.0 cm) below the venipuncture site.
 - o With the 21 gauge needle bevel upward, enter the vein in a smooth continuous motion.
 - o Once blood appears in the butterfly tubing, place the first Vacutainer™ blood tube into the Vacutainer™ holder (Table 33.5). Grasp the flange of the needle holder and push the Vacutainer™ blood tube forward until the butt end of the needle punctures the stopper, exposing the full lumen of the needle.
 - o Make sure the participant's arm is in a flat or downward position while maintaining the Vacutainer™ blood tube below the site when the needle is in the vein. It may be helpful to have the participant make a fist with the opposite hand and place it under the elbow for support. **DO NOT HAVE THE PARTICIPANT MAKE A FIST IN THE HAND OF THE ARM FROM WHICH BLOOD IS TO BE DRAWN.**
 - o Remove the tourniquet after Vacutainer™ blood tube #1 fills. Once the draw has started, do not change the position of a tube until it is withdrawn from the needle. The tourniquet may be reapplied if blood flow is slow without it. If the color of the arm turns red or blue, the tourniquet is applied too tightly. Loosen it and continue.
 - o Keep a constant, slight forward pressure (in the direction of the adapter) on the end of the Vacutainer™ blood tube. This prevents release of the shutoff valve and stopping of blood flow. Do not vary pressure nor reintroduce pressure after completion of the draw.
 - o Fill remaining Vacutainer™ blood tubes in the order specified in Table 33.5.
 - o Fill all Vacutainer™ blood collection tubes as completely as possible, i.e., until the vacuum is exhausted and blood flow ceases. If a Vacutainer™ tube fills only partially, remove the Vacutainer™ blood tube and attach another without removing needle from vein.
 - o When the blood flow into the Vacutainer™ blood tube ceases, remove the Vacutainer™ blood tube from the holder. The shutoff valve covers the point, stopping blood flow until the next Vacutainer™ blood tube is inserted (if necessary).

- o To remove the needle, lightly place clean gauze over venipuncture site. Remove the needle quickly and immediately apply pressure to the site with a gauze pad. Discard needle with its cap into needle box. **DO NOT ATTEMPT TO RECAP NEEDLES.** Have the participant hold the gauze pad firmly for one to two minutes to prevent bruising.
- o If the blood flow stops before collecting all of the Vacutainer™ blood tubes, repeat the venipuncture on the participant beginning with the first unfilled Vacutainer™ blood tube. As always, the tourniquet must never be on for longer than two minutes.

Order of Peripheral Venous Blood Collection

Clinical Blood Collection: This blood is collected and analyzed at each clinical center. Vacutainer™ blood tubes are not provided by the DCC. **Do not send** to Biorepository. For details about clinical blood collection refer to parent study MOP Chapter 30 and Chapter 31.

Center CBC w Diff Results	<u>Note:</u> Lab draw must be no more than 3 months before or after the Form 500 date.
Center Complete Metabolic Panel (CMP) Results	<u>Note:</u> Lab draw <u>must be no more than 3 months before</u> or after the Form 500 date.

- The following clinical bloods done locally: CBC, CMP.
- All other clinical labs: NT-pro BNP, Lipid panel, Inflammation markers, Iron metabolism, Uric acid, Insulin (with calculation of HOMA-IR) will be done by the Biorepository Core

OMICS Blood Collection: Vacutainer™ blood tubes are provided by the DCC.

1. The order of blood draw is shown in Table 233.2
2. Immediately after drawing the samples, gently invert the Vacutainer™ blood collection tubes 10 times to mix the clot activator or anticoagulant with the blood.
3. Place all Vacutainer™ blood tubes on wet ice **except** the Red PAXgene® which will remain at room temperature and can be placed into a biohazard bag for transport.
4. Transport all Vacutainer™ blood tubes to the laboratory. All blood needs to be transported to the lab **within 1 hour of collection.**

Table 233.2 Order of OMICS Blood Draw Collection

Order	Color	Additive	Volume	Core	Transport to Lab
1	Gold	SST	5 ml	CC-PRL	Wet ice
2	Gold	SST	5 ml	CC-PRL	Wet ice
3	Lavender	EDTA	10 ml	OMICS	Wet ice
4	Red PAXgene®	RNA Preservative solution	2.5 ml	Genomic	Room temperature