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Specimen Collection, Processing, Transport, and Storage Protocol

Collection Preparation and Physical Measurements

1. Write the participant name/date of birth (DOB) on the chain-of-custody (COC) and physical examination forms. A blank PDF of the COC and physical examination forms will be emailed to our mobile phlebotomy partners to be printed for each participant prior to the visit.
2. Universal precautions must be taken by all. Phlebotomist, sample collector, and/or any field team member taking physical measurements or handling specimens must wear proper personal protective equipment including powder-free latex or nitrile gloves until all their tasks are complete. **NOTE: If the phlebotomist (or sample collector) is a smoker, they must wear a mask and double gloves (powder-free latex or nitrile) for the entire serum extraction process to prevent contamination of the serum.*
3. Height, weight, pulse, and blood pressure measurements must be recorded on the physical examination form. The study participant number (SP#) and DOB are used to identify the specimens and must be written on all tubes and recorded on all corresponding paperwork.
 - a. Height and weight should be measured with no shoes.
 - b. Blood pressure and heart rate should be measured 3 times for each participant while they are seated. Ideally, there will be two measurements on one arm before the blood draw and one measurement after on the other arm.
 - For NJDOH automatic blood pressure cuff, the wire should line up with the elbow crease.
4. **Specimens must be collected in the lot-screened containers found in the specimen collection kits assembled by NJDOH - substitutions are not allowed.** Each specimen collection kit includes:
 - a. One (1) 100-mL urine collection cup
 - b. Two (2) 5-mL urine aliquot tubes (see Table 1)
 - c. One (1) 6-mL lavender top vacutainer tube for whole blood
 - d. Two (2) 6-mL red top vacutainer tubes for blood that will be used for serum separation
 - e. One (1) 5-mL serum transfer vial
 - f. Two (2) transfer pipettes (one each for urine and serum)
5. Serum specimens **must** be processed at a minimum of 1 hour and maximum of 1.5 hours after collection to ensure specimen integrity, as described in the specimen processing section below.

Specimen Collection and Processing

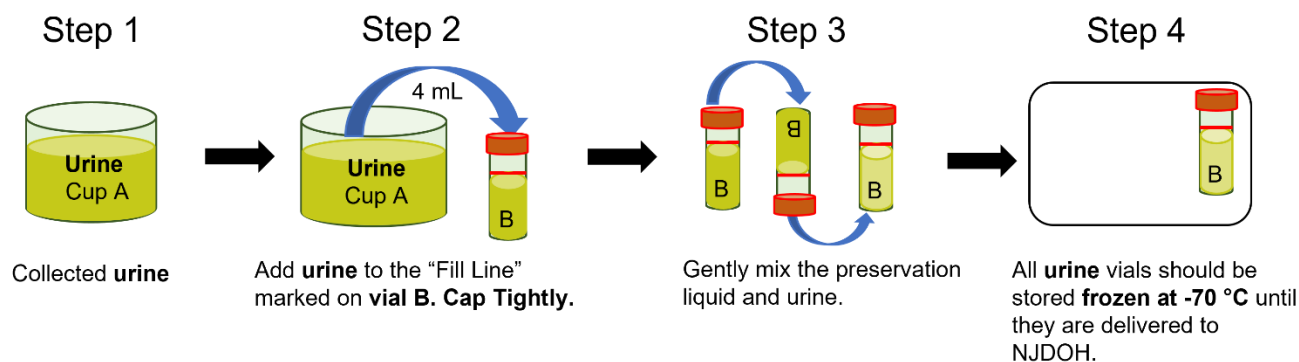
Urine

Table 1. Urine Processing Information.

Number of Containers	Container Type	Specimen Code
1	100 mL Cup	Cup A
1	5 mL Vial	Vial B (Preserved)
1	5 mL Vial	Vial C

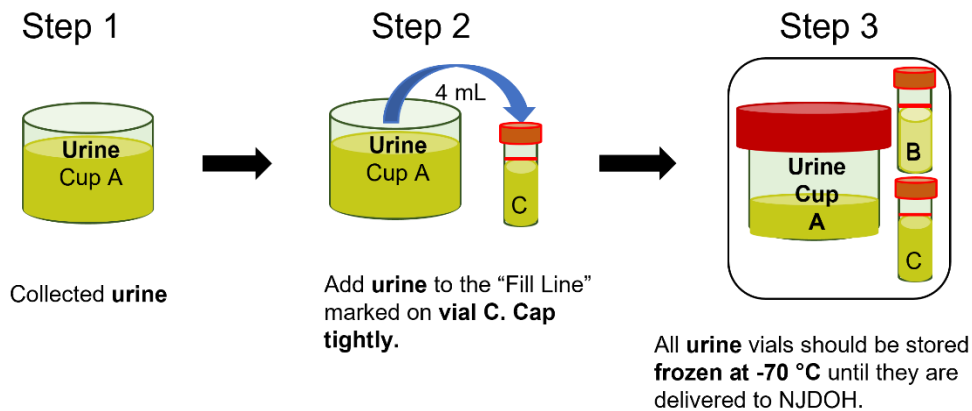
1. Collect a minimum of 30 mL urine into the 100 mL urine cup (A).
 - When scheduling visit, advise participant to drink water before the exam at least 30 minutes prior to the scheduled visit.
2. Transfer urine from the 100 mL collection container using the plastic transfer pipette to ~4 mL to Vial B (preserved). Cap the vial tightly. See NJHANES Urine Processing I diagram below.
3. Repeat for Vial C. See NJHANES Urine Processing II diagram below.
4. Immediately freeze urine samples (A-C) in a -70 °C freezer (keep on ice if in the field, then freeze immediately upon return) for long-term storage. For NJDOH collections, transport on provided icepacks to NJDOH and proceed to sample log in.

NJHANES Urine Processing (I)



*Note: Urine Tubes B & C can be easily differentiated with labels of 'UrineP' and just 'Urine', respectively.

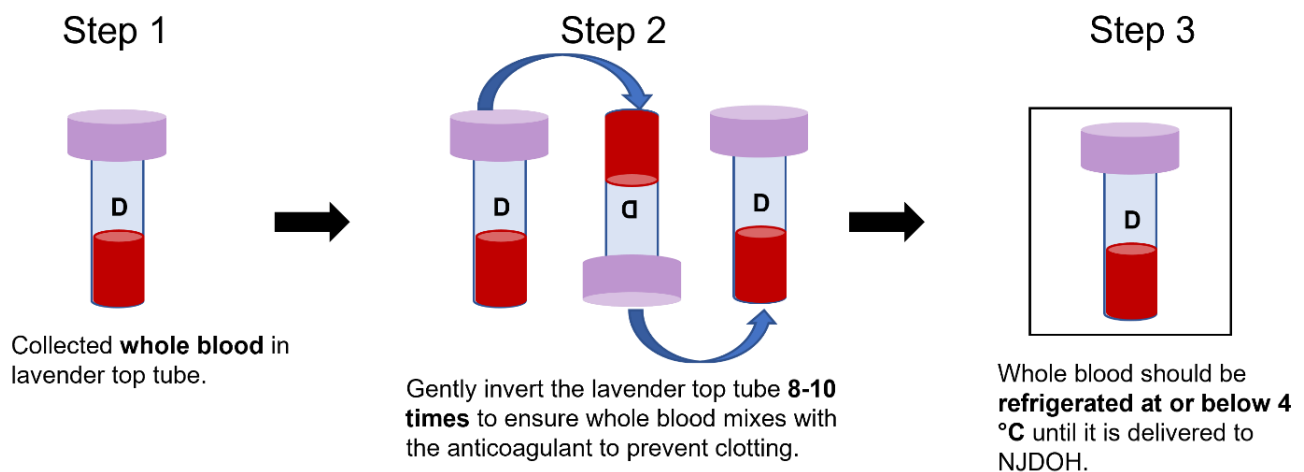
NJHANES Urine Processing (II)



Whole Blood

1. Take out the lavender top vacutainer tube labeled "D" and collect ~5 mL whole blood.
2. After the whole blood is collected, gently mix by inverting the tube 8-10 times. See NJHANES Whole Blood Processing diagram below.
3. Store the specimen upright in a cooler to keep refrigerated at 4 °C or below during transport. Continue to store at 4 °C or below until they are delivered to NJDOH.

NJHANES Whole Blood Processing

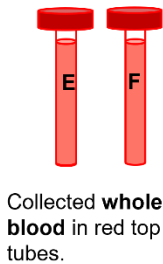


Serum

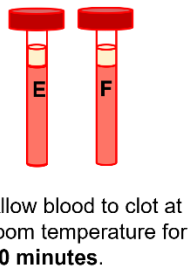
1. Take out the red top blood collection vacutainers labeled “E” and “F” and the serum transfer vial labeled “Vial G” (and “H”, if needed).
2. Fill the red top tubes, then gently invert 5-6 times to mix well. See NJHANES Serum Processing diagram below.
3. Let the blood clot in the red top vacutainer tubes at room temp for a minimum of 1 hour and maximum of 2 hours.
4. After the clotting period, spin down the clotted blood at room temperature for 15 minutes using a centrifuge (1000 G – 2400 G) to separate the serum from the clotted blood. Carefully transfer the separated serum (pale yellow layer on top) from each red top tube into serum transfer Vial E. **NOTE: Never recentrifuge specimens. Never touch or introduce clotted blood into the serum storage container.**
5. Freeze samples (Vials E, F, and G [and H, if needed]) at -70 °C (keep on ice if in the field, then freeze immediately upon return) until they are delivered to NJDOH.

NJHANES Serum Processing

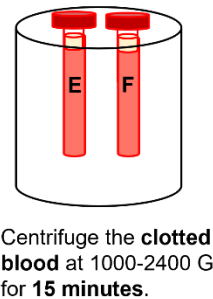
Step 1



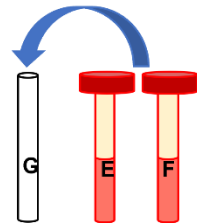
Step 2



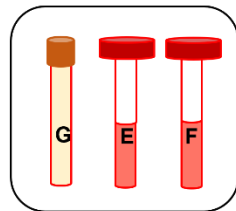
Step 3



Step 4

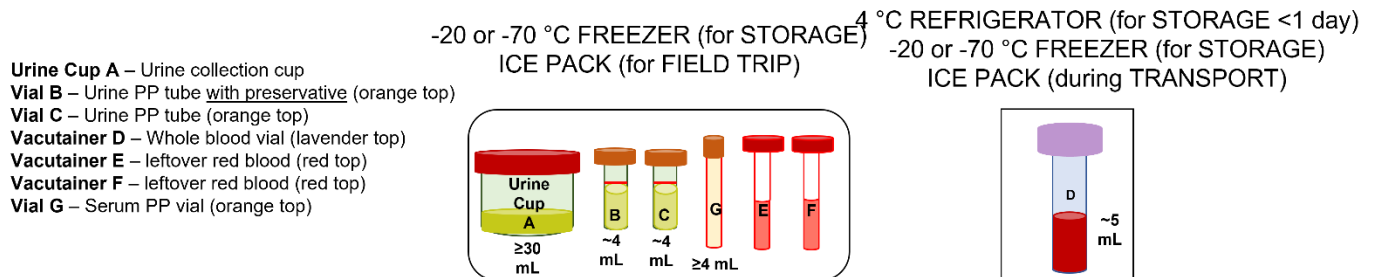


Step 5



Specimen Transport and Storage Conditions

NJHANES Specimen Storage



1. All specimens must be kept <4 °C in the field once processed. Urine and serum specimens must be stored at -70 °C or below long term and whole blood must be stored at or below 4 °C.
2. Specimens must be placed upright in the grid box provided by NJDOH for transport.
3. Specimens should be transported or stored only using NJDOH-provide ice packs to avoid contamination.
4. The completed paperwork must accompany each shipment for each set of specimens:
 - a. Chain-of-Custody form with participant name/DOB
 - b. Physical Exam form with participant name/DOB
5. Contact NJDOH to coordinate specimen transport to NJDOH.
6. For shipping via the courier service:
 - a. NJDOH will reach out to coordinate specimen pick up time by the courier.
 - b. Place the specimens in the shippers provided by NJDOH. Follow the packing instructions to pack the specimens.
 - c. Sign the COC form where indicated, place the hard copy of the paperwork in a Ziploc bag, and ship it with the specimens.
 - d. Add ice to the container and hand the package to the courier. Email the NJDOH to let them know the package was picked up by the courier.
7. Contact information for logistical issues will be provided.
8. Contact information for specimen technical issues will be provided.

Specimen Acceptance and Rejection Criteria

1. Specimens must meet the total minimum volume requirements as shown in the table below.

Matrix	Total Minimum Required Volume (mL)
Whole Blood	2.5
Serum	3.5
Urine	20

2. Serum specimens must be free from clots.
3. ANY SPECIMEN NOT COLLECTED IN NJDOH CONTAINERS MUST BE REJECTED.
4. Chain-of-custody forms must accompany each set of specimens during transport.