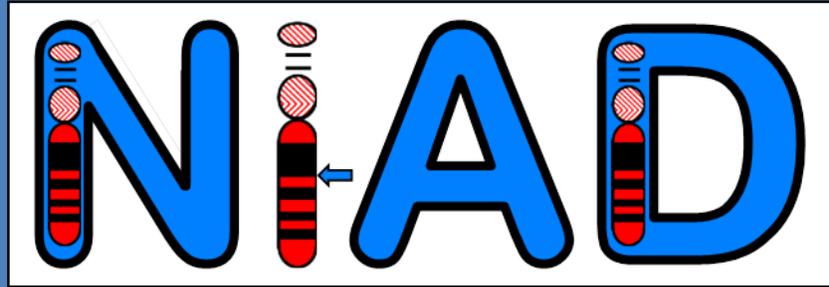




NiAD MANUAL OF PROCEDURES UPDATE:

V03.2020

| Section | Change |
|---------------------|---|
| Document Footer | The version date was updated for this amendment. |
| Title Page | Title page updated to reflect NCRAD name change. |
| Throughout document | 1. Updated fax number and room number throughout. |



**Neurodegeneration in Aging Down Syndrome: A Longitudinal
Study of Cognition and Biomarkers of Alzheimer's Disease**

in collaboration with

**The National Centralized Repository for Alzheimer's Disease
and Related Dementias (NCRAD)**

Blood-Based Biospecimens

Manual of Procedures

Version 4.3

March, 2020



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1.0 ABBREVIATIONS

| | |
|--------|---|
| ABC-DS | Alzheimer's Biomarker Consortium – Down Syndrome |
| AD | Alzheimer's Disease |
| ADDS | Alzheimer's Disease in Down Syndrome |
| ATRI | Alzheimer's Therapeutic Research Institute |
| CSF | Cerebrospinal Fluid |
| DNA | Deoxyribonucleic Acid |
| DS | Down Syndrome |
| EDTA | Ethylene Diamine Tetra-acetic Acid |
| IATA | International Air Transport Association |
| IUGB | Indiana University Genetics Biobank |
| NaHep | Sodium Heparin (Green-Top) Blood Collection Tube (10 ml) |
| NCRAD | National Centralized Repository for Alzheimer's Disease and Related Dementias |
| NiAD | Neurodegeneration in Aging Down Syndrome: A Longitudinal Study of Cognition and Biomarkers of Alzheimer's Disease |
| PBMC | Peripheral Blood Mononuclear Cell |
| RBC | Red Blood Cells |
| RCF | Relative Centrifugal Force |
| RPM | Revolutions Per Minute |
| SST | Serum Separator Tube |

2.0 PURPOSE

The collection of blood-based biofluids is an important part of the Neurodegeneration in Aging Down Syndrome (NiAD) study. These samples will be used to:

- Identify critical factors that link A β deposition to neurodegeneration and, ultimately, dementia
- Define biomarkers for the above factors
- Set a foundation for an efficient transition from this biomarker study to a therapeutic trial to combat AD in DS augmented by biomarker outcomes

The purpose of this manual is to provide study staff (PIs, study coordinators, phlebotomists) at the various study sites with instructions for collection and submission of blood-based biological samples for NiAD study visits. It includes instructions for blood-based biospecimen submission to NCRAD located in Indianapolis at Indiana University.

The following samples will be sent to NCRAD:

- Serum – (DS participants ONLY)
- Plasma – (DS participants AND sibling controls)
- Buffy Coat - (DNA extraction) – (DS participants AND sibling controls)

Additional samples collected but not shipped to NCRAD:

- CSF (Please see the ABC-DS Lumbar Puncture Manual of Procedures for details)

This manual includes instructions for collection of blood, fractionation of blood from collection tubes, aliquoting, labeling, storage prior to shipping, and shipping to NCRAD.

These procedures are relevant to all study personnel responsible for processing blood specimens entering NCRAD.



3.0 NCRAD INFORMATION

3.1 NCRAD Contacts

Tatiana Foroud, PhD, NCRAD Leader

Phone: 317-274-2218

Kelley Faber, MS, CCRC, Project Manager

Phone: 317-274-7360

Email: kelfaber@iu.edu

Colleen Mitchell, Laboratory Manager

Phone: 317-278-9016

Email: mitchechm@iu.edu

General NCRAD Contact Information

Phone: 1-800-526-2839

Fax: 317-321-2003

Email: alzstudy@iu.edu

Website: www.ncrad.org

ADDS-NiAD Study Specific Webpage:

https://www.ncrad.org/resource_adds_niad.html

Kristi Wilmes, MS, CCRP Study Coordinator

Phone: 317-274-7546

Email: wilmesk@iu.edu

Sample Shipment Mailing address

NCRAD

Indiana University School of Medicine

351 West 10th Street

TK-217

Indianapolis, IN 46202



3.2 Hours of Operation

Indiana University business hours are from 8 AM to 5 PM Eastern Time, Monday through Friday.

Frozen samples must be shipped **Monday-Wednesday only**.

Check weather report to make sure impending weather events (blizzards, hurricanes, etc.) will not affect the shipping or delivery of the samples.

3.3 Holiday Schedules

- Please note that courier services may observe a different set of holidays. Please be sure to verify shipping dates with your courier prior to any holiday.

3.4 Holiday Observations

| Date | Holiday |
|--------------------------------------|-----------------------------|
| January 1 | New Year's Day |
| 3 rd Monday in January | Martin Luther King, Jr Day |
| 4 th Monday in May | Memorial Day |
| July 4 | Independence Day (observed) |
| 1 st Monday in September | Labor Day |
| 4 th Thursday in November | Thanksgiving |
| 4 th Friday in November | Friday after Thanksgiving |
| December 25 | Christmas Day |

Please note that between December 24th and January 2nd, Indiana University will be open Monday through Friday for essential operations **ONLY** and will re-open for normal operations on January 2nd. If at all possible, biological specimens for submission to Indiana University should **NOT** be collected and shipped to Indiana University after the second week in December. Should it be necessary to ship blood samples for DNA extraction to Indiana University during this period, please contact the Indiana University staff before December 20th by e-mailing alzstudy@iu.edu, so that they can arrange to have staff available to process incoming samples.

Please see: https://ncrad.org/holiday_closures.html for additional information.

4.0 NCRAD LABORATORY COLLECTION

4.1 Site Required Equipment

The following materials and equipment are necessary for the processing of specimens at the collection site and are to be **supplied by the local site**:

- Personal Protective Equipment: lab coat, nitrile/latex gloves, safety glasses
- Tourniquet
- Alcohol Prep Pad
- Gauze Pad
- Bandage
- Butterfly needles (21 gauge) and hub
- Microcentrifuge tube rack
- Sharps bin and lid
- Wet Ice Bucket

In order to process samples consistently across all projects and ensure the highest quality samples possible, project sites must have access to the following equipment:

- Centrifuge capable of ≥ 2000 rcf with refrigeration to 4°C
- -80°C Freezer

In order to ship specimens, you must provide:
Dry ice (about approximately 30-45 lbs per shipment)

4.2 BIOSPECIMENS COLLECTION SCHEDULES (SPECIMENS SENT TO NCRAD ONLY)

NiAD Blood-Based Biomarker Collection Schedule for DS Participants:

| | Baseline | 16-Month | 32-Month | 48-Month |
|-------------|----------|----------|----------|----------|
| Serum | X | X | X | ** |
| DNA | X | X* | X | ** |
| Plasma | X | X | X | ** |
| Karyotyping | X | | | |

*DNA from the 16-Month Visit (University of Pittsburgh ONLY) will not be shipped to NCRAD, but maintained at the site.

**No Blood-Based Biomarkers are collected at the 48-month visit

Whole blood is collected in two different types of tubes (gold top Serum Separator tubes and lavender top EDTA tube) for shipment to NCRAD. These two tubes are processed locally into serum, plasma, and buffy coat fractions; they are then aliquoted, frozen at the study site, and shipped to NCRAD.



NiAD Blood-Based Biomarker Collection Schedule for Sibling Controls:

| | Baseline | 16-Month | 32-Month | 48-Month |
|--------|----------|----------|----------|----------|
| DNA | X | | | |
| Plasma | X | | | |

Whole blood is collected in one tube, a lavender top EDTA tube, for processing and shipment to NCRAD. This tube is processed locally into plasma and buffy coat fractions; it is aliquoted, frozen at the study site, and shipped to NCRAD. This tube is collected at baseline only for sibling controls.

Consent forms must specify that any biological samples and de-identified clinical data may be shared with academic and/or industry collaborators through NCRAD. A copy of the consent form for each subject should be kept on file by the site investigator.

Frozen samples are to be submitted according to the shipping methods outlined in [Section 8.1](#). Guidelines for the processing, storage location, and timing of sample collection are listed in the tables below.

4.2.1 RE-DRAW INSTRUCTIONS AND TIMEFRAMES

Sample Collection-Blood eCRF is a log form. Select ‘*Add a new record*’ to enter a record. Enter one record per Date of Collection and specify samples collected. At least one sample type must be marked as collected on this date to successfully submit the form.

If a re-draw is necessary and occurs BETWEEN TWO VISITS, add a new record in the visit PRIOR to the re-draw timeframe, making sure to include the re-draw Date of collection and Kit Number. If a sample was missed during a regularly scheduled visit, but a sample was collected PRIOR to NEXT scheduled visit, enter in the EDC as a re-draw. Also, provide reason for re-draw in the Comments section.

For NiAD, the re-draw timeframe is as follows:

- For baseline, the re-draw timeframe will be up to 0-3 months after the baseline visit.
- For the 16-month visit, the re-draw timeframe will be 13-19 months after the baseline visit.
- For the 32-month visit, the re-draw timeframe will be 29-35 months after the baseline visit.



- For the 48-month visit, the re-draw timeframe will be 45-51 months after the baseline visit.

4.3 BIOSPECIMEN COLLECTION CHART

4.3.1 Blood Collection for DS Participants

| Sample Type | Tube Type | Number of Tubes Supplied in Kit | Processing/ Aliquoting | Tubes to NCRAD |
|---|--|---------------------------------|--|----------------|
| Whole blood for isolation of serum | Serum Separator (Gold-Top) Blood Collection Tube (5 ml) | 2 | N/A | N/A |
| | SERUM: 0.5 ml cyrovials | 21 | 0.25 ml serum aliquot per 0.5 ml siliconized cyrovial (clear cap with RED sticker) | 16-21 |
| Whole blood for isolation of plasma & buffy coat (for DNA extraction) | EDTA (Lavender-Top) Blood Collection Tube (10 ml) | 1 | N/A | N/A |
| | PLASMA: 0.5 ml cyrovials | 21 | 0.25 ml plasma aliquot per 0.5 ml siliconized cryovial (clear cap with LAVENDER sticker) | 16-21 |
| | BUFFY COAT: 2.0 ml cryovial | 1 | 1 ml buffy coat aliquot per 2.0 ml cryovial (BLUE CAP) | 1 |
| Whole blood for Karyotyping | Sodium Heparin (Green-Top) Blood Collection tube (10 ml) | 1 | N/A | N/A |

If a sample is not obtained at a particular visit, this should be recorded in the notes section of the **Biological Sample and Shipment Notification Form** (see [Appendix B](#)). Submit a copy to NCRAD with a reason provided for the omission.

4.3.2 Blood Collection for Sibling Controls

| Sample Type | Tube Type | Number of Tubes Supplied in Kit | Processing/ Aliquoting | Tubes to NCRAD |
|---|---|---------------------------------|--|----------------|
| Whole blood for isolation of plasma & buffy coat (for DNA extraction) | EDTA (Lavender-Top) Blood Collection Tube (10 ml) | 1 | N/A | N/A |
| | PLASMA: 0.5 ml cryovials | 21 | 0.25 ml plasma aliquot per 0.5 ml siliconized cryovial (clear cap with LAVENDER sticker) | 16-21 |
| | BUFFY COAT: 2.0 ml cryovial | 1 | 1 ml buffy coat aliquot per 2.0 ml cryovial (BLUE CAP) | 1 |

If a sample is not obtained at a particular visit, this should be recorded in the notes section of the **Biological Sample and Shipment Notification Form** (see [Appendix B](#)). Submit a copy to NCRAD with a reason provided for the omission.

5.0 SPECIMEN COLLECTION KITS, SHIPPING KITS, AND SUPPLIES

NCRAD will provide: 1) Blood sample collection kits for research specimens to be stored at NCRAD; 2) CSF collection kits including Lumbar Puncture (LP) trays, the CSF Supplemental Supply Kit and the CSF Shipping Supply Kit; and 3) clinical lab supplies (with the exception of dry ice and equipment supplies listed in [Section 4.1](#)). These materials include blood tubes, pipettes, pipette tips, LP trays (when applicable), boxes for serum/plasma/buffy coat/CSF aliquots, as well as partially completed shipping labels to send materials to NCRAD. Kit Number Labels, Site and NiAD ID Labels, Collection and Aliquot Tube Labels will all be provided by NCRAD. Details regarding the CSF Kits are found in the ABC-DS LP Manual of Procedures. Collection and Aliquot Tube Labels will be pre-printed with study information specific to the type of sample being drawn. Ensure that all tubes are properly labeled during processing and at the time of shipment according to [Section 6.1](#).

5.1 Specimen Collection Kit Contents

Collection kits contain the following (for each subject) and provide the necessary supplies to collect samples from a given subject. Do not replace or supplement any of the tubes or kit components provided with your own supplies unless you have received approval from the NCRAD Study team to do so. *Please store all kits at room temperature until use.*

NiAD Blood Kit for DS Participants

| Quantity | NiAD Blood-Based Kit Components |
|----------|---|
| 1 | EDTA (Lavender-Top) Blood Collection Tube (10 ml) |
| 2 | Serum Separator (Gold-Top) Blood Collection Tube (5 ml) |
| 1 | 15 ml conical |
| 21 | Siliconized cryovial tube (0.5 ml) with clear cap with lavender sticker |
| 21 | Siliconized cryovial tube (0.5 ml) with clear cap with red sticker |
| 1 | Cryovial tube (2.0) with blue cap |
| 1 | Disposable graduated transfer pipette |
| 46 | Pre-printed Collection and Aliquot Tube Label |
| 4 | Pre-printed Kit Number Label |
| 4 | Labels for handwritten Site and NiAD ID |
| 2 | Microcentrifuge tube box (holds up to 25 microcryovials) |

NiAD Sibling Control Blood Kit

| Quantity | NiAD Blood-Based Kit Components |
|----------|---|
| 1 | EDTA (Lavender-Top) Blood Collection Tube (10 ml) |
| 21 | Siliconized cryovial tube (0.5 ml) with clear cap with lavender sticker |
| 1 | Cryovial tube (2.0) with blue cap |
| 1 | Disposable graduated transfer pipette |
| 23 | Pre-printed Collection and Aliquot Tube Label |
| 3 | Pre-printed Kit Number Label |
| 3 | Labels for handwritten Site and NIAD ID |
| 1 | Microcentrifuge tube box (holds up to 25 microcryovials) |

NiAD Frozen Blood Shipping Supply Kit

| Quantity | Frozen Shipping Kit Components for Blood-Based Biomarkers |
|----------|---|
| 5 | Plastic Biohazard bag with absorbent sheet |
| 1 | FedEx return airbill and pouch |
| 1 | Shipping box/Styrofoam container |
| 1 | Warning label packet with dry ice sticker |

Blood Supplemental Supply Kit

| Quantity | Blood-Based Supplemental Supply Kit Components |
|----------|---|
| 10 | Siliconized cryovial tube (0.5 ml) with clear cap with lavender sticker |
| 10 | Siliconized cryovial tube (0.5 ml) with clear cap with red sticker |
| 5 | Cryovial tube (2.0 ml) with blue cap |
| 5 | 15 ml conical |
| 5 | Microcentrifuge tube box (holds up to 25 microcryovials) |
| 5 | Disposable graduated transfer pipette |
| 5 | EDTA (Lavender-Top) Blood Collection Tube (10 ml) |
| 10 | Serum Separator (Gold-Top) Blood Collection Tube (5 ml) |
| 20 | Sodium Heparin (Green-Top) Blood Collection Tube (10 ml) |
| 10 | Labels for handwritten Site and NIAD ID |

Individual Supplies

| Quantities | Items Available upon request within the NCRAD kit module. |
|------------|--|
| By Request | 25 cell cryobox |
| By Request | Siliconized Cryovial tube (0.5) with clear cap with lavender sticker |
| By Request | Siliconized Cryovial tube (0.5) with clear cap with red sticker |
| By Request | Cryovial tube (2 ml) with blue cap |

| | |
|------------|--|
| By Request | FedEx return airbill |
| By Request | 15 mL conical tube |
| By Request | Small IATA shipping box for ambient shipping |
| By Request | Shipping container for dry ice shipment (shipping and Styrofoam box) |
| By Request | Plastic biohazard bag |
| By Request | Disposable graduated transfer pipette |
| By Request | EDTA (Lavender-Top) Blood Collection Tube (10 ml) |
| By Request | Serum Separator (Gold-Top) Blood Collection Tube (5 ml) |
| By Request | Sodium Heparin (Green-Top) Blood Collection Tube (10 ml) |
| By Request | Warning label packet |
| By Request | Fine Point permanent markers |
| By Request | Site and NIAD ID Labels |

5.2 Kit Supply to Study Sites

Each individual site will be responsible for ordering and maintaining a steady supply of kits from NCRAD. We advise sites to keep a supply of each kit type available. Be sure to check your supplies and order additional materials before you run out or supplies expire so you are prepared for study visits. Please go to [ADDs-NiAD Kit Request System](#) to request additional kits and follow the prompts to request the desired supplies. Options include ordering a specific number of kits; we are also including the option of simply ordering the desired amount of extra supplies.

Please allow **TWO weeks** for kit orders to be processed and delivered.

6.0 BLOOD COLLECTION AND PROCESSING PROCEDURES

Important Note

In order to ensure the highest quality samples are collected, processed, and stored, it is essential to follow the specific collection, processing, and shipment procedures detailed in the following pages. Please read the following instructions first before collecting any specimens. Have all your supplies and equipment out and prepared prior to drawing blood. Draw blood in the following order:

1. Serum Separator (Gold-Top) Blood Collection Tube (5 ml) for Serum x 2
(DS Participants ONLY)
2. EDTA (Lavender-Top) Blood Collection Tube (10 ml) for DNA and Plasma
3. Sodium Heparin (Green-Top) Blood Collection Tube (10 ml) for Karyotyping
(Baseline ONLY; DS Participants ONLY)

SPECIFIC INSTRUCTIONS FOR COLLECTION AND PROCESSING OF EACH SAMPLE ARE DETAILED ON THE FOLLOWING PAGES.

6.1 Labeling Samples

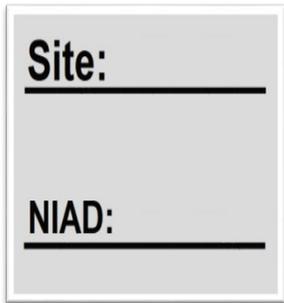
- **Label Type Summary**
1. Kit Number Label
 2. Collection and Aliquot Tube Label
 3. Site and NIAD ID Label



The **Kit Number Labels** do not indicate a specimen type, but are affixed on the Biological Sample and Shipment Notification Forms and on specific packing materials.



The **Collection and Aliquot Tube Labels** for blood derivatives and CSF are placed on all collection and aliquot tubes.



The **Site and NIAD ID Labels** are placed on all collection tubes, both blood and CSF.

****Important Note****
Each collection tube will contain two labels: the Collection and Aliquot Tube Label and the Site and NIAD ID Label. Be sure to place labels in the same configuration consistently among tubes, with the barcoded label near the top of the tube and the handwritten Site and NIAD ID label.

****Important Note****
The label for the sodium heparin (green-top) blood collection tube (10 ml) to be drawn for karyotyping will not be included in the NCRAD provided blood kits. The labels for these tubes should be created on site with information requested by the local lab.



Serum Separator (Gold-Top) Blood Collection Tube (5 ml)

Drawn for DS Participants ONLY

Collection and Cryovial Tube Label

Site and NIAD ID Label

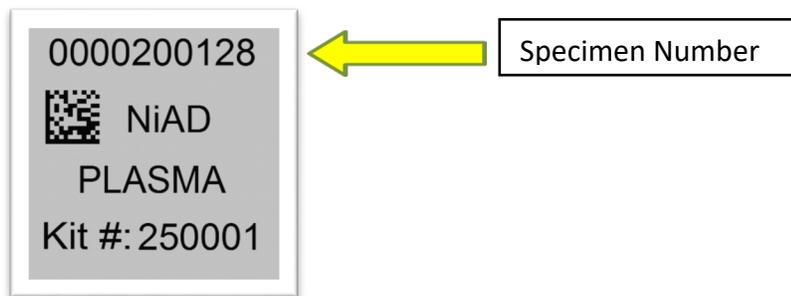


EDTA (Lavender-Top) Blood Collection Tube (10 ml)

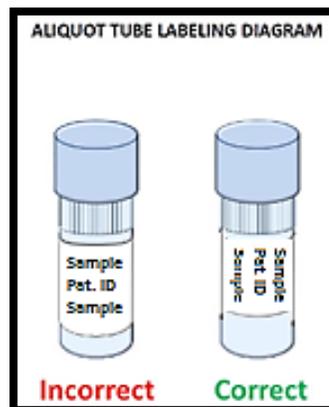
Drawn for DS Participants and Sibling Controls

In order to ensure the label adheres properly and remains on the tube, please follow these instructions:

- Place blood collection and aliquot labels on **ALL** collection and aliquot tubes **BEFORE** sample collection, sample processing, or freezing. This should help to ensure the label properly adheres to the tube before exposure to moisture or different temperatures.
- Place cryovials in numerical order based on the specimen number, located at the top of the label. This ensures that no aliquot is misplaced or lost during the shipment process (See Plasma Collection and Aliquot Tube Label on next page.)



- Using a fine point permanent marker, fill-in and place the Site and NIAD ID Labels on the collection tubes only (EDTA, SST) **BEFORE** sample collection, processing, or freezing. These labels are in addition to the Collection and Aliquot Tube Labels. **DO NOT** place Site and NIAD ID labels on any cryovials.
- The Collection and Aliquot Tube Labels contain a 2D barcode on the left hand side of the label. Place this barcode toward the tube cap.
- Place label **horizontally** on the tube (wrapped around sideways if the tube is upright) and **just below the ridges** of the aliquot tubes (see labeling diagram below).
- Take a moment to ensure the label is **completely adhered** to each tube. It may be helpful to roll the tube between your fingers after applying the label.



- If there are any unused cryovials, please do not send the empty cryovials to NCRAD. These unused cryovials (ensure labels are removed) can be saved as part of a supplemental supply at your site or the cryovials can be disposed of per your site's requirements.

6.2 Video List

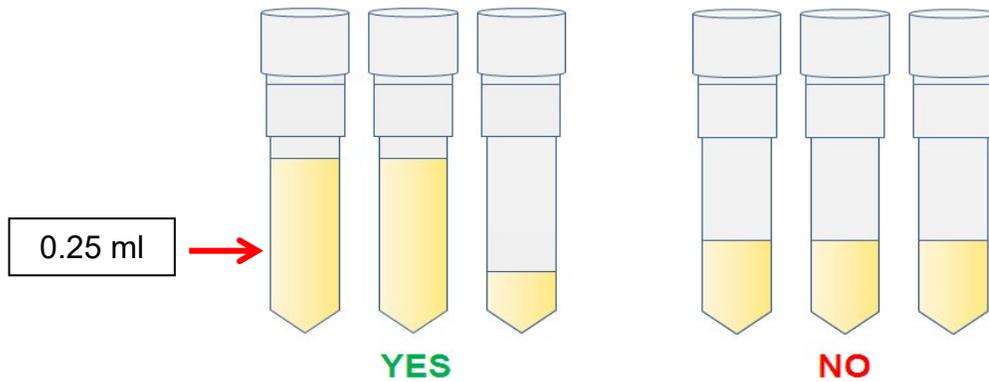
- The following training videos are available to assist you with the specimen processing, aliquoting, and shipping processes. The videos are available at:

https://ncrad.org/resource_adds_niad.html.

- NiAD MOP Training
- Plasma and Buffy Coat Processing and Aliquoting
- Serum Processing and Aliquoting
- Frozen Shipping

6.3 Filling Aliquot Tubes (Plasma and Serum)

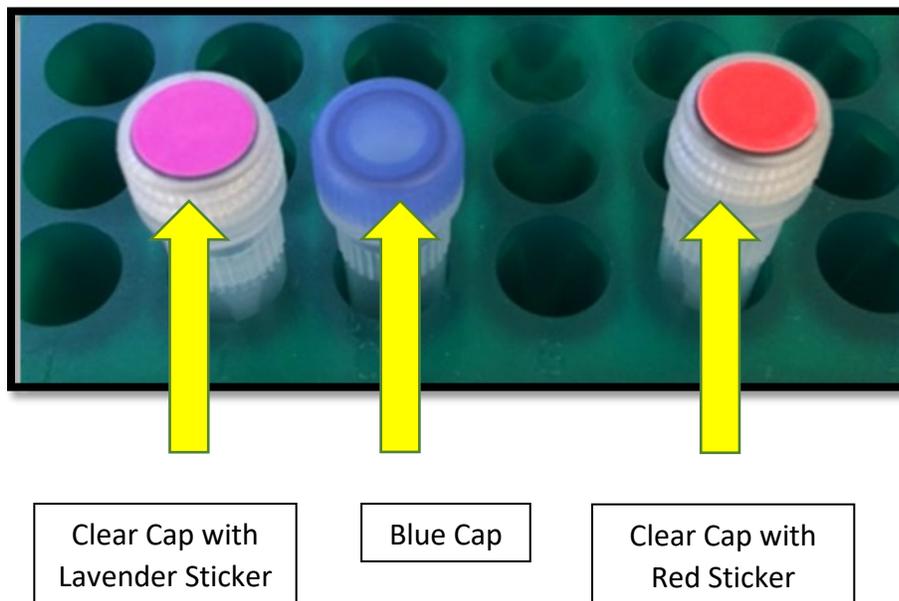
In order to ensure that NCRAD receives a sufficient amount of sample for processing and storage, and to avoid cracking of the tubes prior to shipment, each cryovial should be filled to 0.25 milliliters (see picture below) with the respective biological material after processing is completed (refer to detailed processing instructions for average yield per sample). Over-filled tubes may burst once placed in the freezer, resulting in a loss of that sample. If there is biologic material remaining that will not fill a subsequent cryovial, that remaining amount should still be included and shipped to NCRAD. Essentially, all material should be shipped to NCRAD, ensuring maximum amount in as many cryovials as will allow after processing the sample. You do not have to fill all cryovial tubes provided; you should attempt to fill as many tubes as possible with 0.25 ml of sample. For example, if 3.6 ml of sample is obtained, you should fill 14 cryovial tubes each with 0.25 ml, and one additional cryovial tube with the remaining 0.1 ml.



Please note: It is critical for the integrity of the samples that study staff note if an aliquot tube contains a residual volume (anything under 0.25 ml). Please record the specimen number and volume of the residual aliquot on the Biological Sample and Notification Form.

To assist in the preparation and aliquoting of samples, colored caps and cap stickers are used for the cryovial tubes. The chart below summarizes the association between cap color and type of cryovial.

| Cap Color | Sample Type |
|---------------------------------|-------------|
| Clear Cap with Lavender Sticker | Plasma |
| Blue Cap | Buffy Coat |
| Clear Cap with Red Sticker | Serum |



6.4 Serum Separator (Gold-Top) Blood Collection Tube (5 ml) for Serum

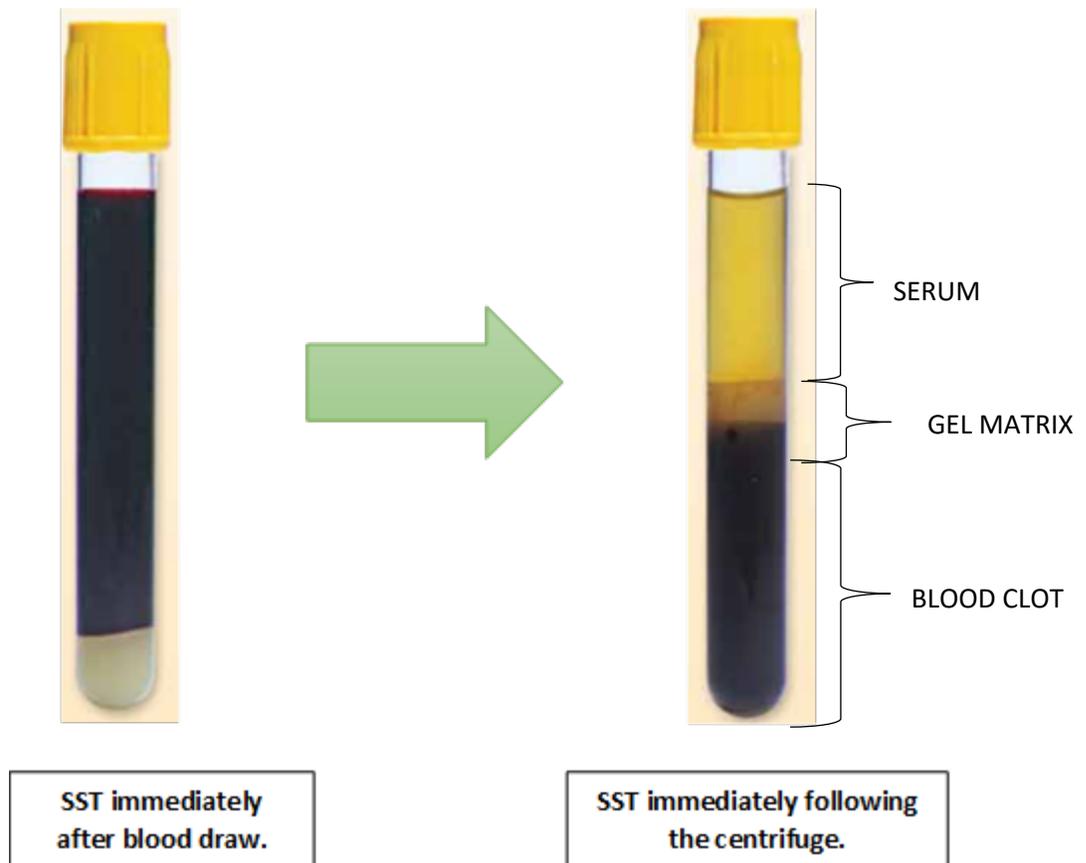
Whole Blood Collection for Isolation of Serum: Serum Separator (Gold-Top) Blood Collection Tube (5 ml) (for processing of serum aliquots).

1. Set centrifuge to 4°C to pre-chill before use.
2. Place completed Site and NIAD ID Label and Collection and Aliquot “**SERUM**” Tube Labels on the Serum Separator (Gold-Top) Blood Collection Tubes (2 x 5 ml). Place pre-printed Collection and Aliquot “**SERUM**” Tube Labels on the (21) 0.5 ml siliconized cryovial tubes with clear caps and red stickers.
3. Using a blood collection set and a holder, collect blood into **Serum Separator (Gold-Top) Blood Collection Tubes (2 x 5 ml)** using your institution's recommended procedure for standard venipuncture technique

The following techniques shall be used to prevent possible backflow:

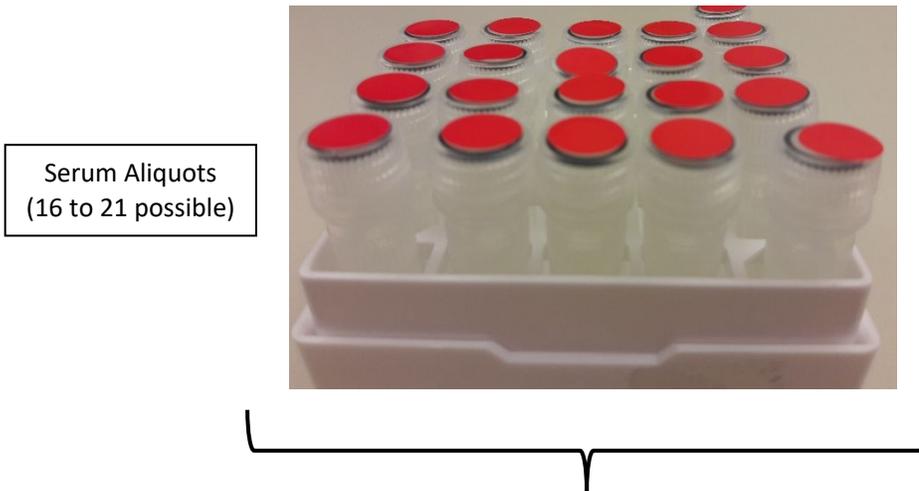
- a. Place donor's arm in a downward position.
 - b. Hold tube in a vertical position, below the donor's arm during blood collection.
 - c. Release tourniquet as soon as blood starts to flow into tube.
 - d. Make sure tube additives do not touch the stopper or the end of the needle during venipuncture.
4. Allow at least 10 seconds for a complete blood draw to take place in each tube. **Ensure that the blood has stopped flowing into each tube before removing the tube from the holder.** The tube with its vacuum is designed to draw 5 ml of blood into the tube.
 - a. If complications arise during the blood draw, please note the difficulties on the ‘Biological Sample and Shipment Notification Form’. Do not attempt to draw an additional SST at this time. Process blood obtained in existing SST tube.
 5. **CRITICAL STEP: Immediately after blood collection, gently invert/mix (180 degree turns) each tube 5 times.**
 6. **CRITICAL STEP: Allow blood to clot at room temperature by placing it upright in a vertical position in a tube rack for 30 minutes. Serum samples need to be spun, aliquoted, and placed in the freezer within 2 hours from the time of collection.**

7. After 30 minutes of clotting, centrifuge the collection tube for 10 minutes at 2000 RCF (x g) at 4°C. **It is critical that the tube be centrifuged at the appropriate speed to ensure proper serum separation (see worksheet in [Appendix A](#) to calculate RPM.**
 - Equivalent rpm for spin at 2000 x g
 - While centrifuging, remember to record all times, temperatures and spin rates on the Biological Sample and Shipment Notification Form [Appendix B](#).
 - Serum samples need to be spun, aliquoted, and placed in the freezer within 2 hours from the time of collection.
 - Record time aliquoted on the Biological Sample Shipment and Notification Form



8. Remove the serum by tilting the tube and placing the pipette tip along the lower side of the wall. Transfer serum from both gold top Serum Separator tubes into the single 15ml conical tube. Mix the serum by gently inverting the conical tube 3-4 times.
9. Using a pipette, transfer serum from the 15 ml conical tube, into the pre-labeled siliconized cryovials with the red stickers on the caps. Aliquot 0.25 ml

per cryovial (total vials=16-21 with 0.25 ml each). One Gold-Top tube should yield, on average, 5 ml of blood serum. Between both of the Gold-Top tubes, there should be an average of 10 ml of serum, for a total of 16-21 0.5 ml aliquot cryovial tubes per subject with 0.25 ml per cryovial tube. Be sure to only place **serum** in siliconized cryovials labeled with the "SERUM" label and red stickers on the caps. If there is extra serum left, use 1 extra cryovial provided for another <0.25 ml aliquot of serum and label as appropriate. **If a residual aliquot (<0.25 ml) is created, document the sample number and volume on the Biological Sample and Shipment Notification Form.**



Close up view of 0.5 ml
Siliconized Cryovial

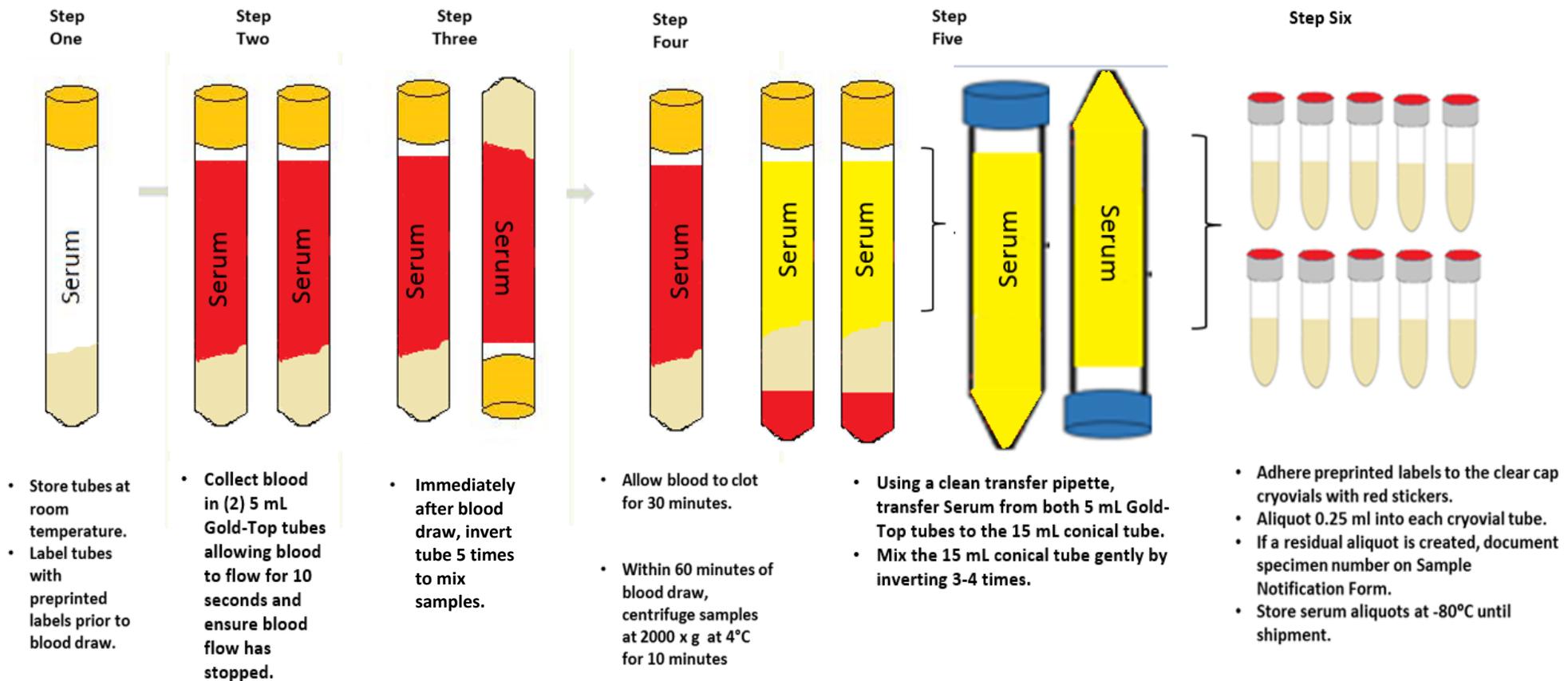


10. Place the labeled cryovials in the 25 cell cryobox and place on dry ice. Transfer to **-80°C Freezer when possible**. Store all samples at **-80°C until shipped** to NCRAD on dry ice. Record time aliquots placed in freezer and storage temperature of freezer on Biological Sample and Shipment Notification Form.



11. Dispose of collection tube with gel matrix and red blood cells at the bottom of the tube and empty 15ml conical tube according to your site's guidelines for disposing of biomedical waste.

Serum Preparation (5 ml Gold-Top Tube)



6.5 EDTA (Lavender-Top) Blood Collection Tube (10 ml) for Plasma and Buffy Coat

Whole Blood Collection for Isolation of Plasma and Buffy Coat: EDTA (Lavender-Top) Blood Collection Tube (10 ml) (for processing of plasma aliquots and buffy coat aliquot).

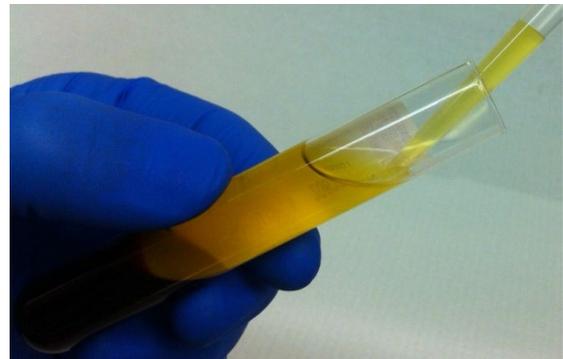
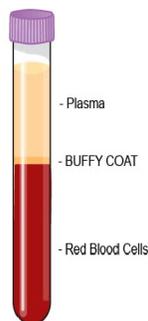
1. Set centrifuge to 4°C to pre-chill before use.
2. Place completed Site and NIAD ID Label and pre-printed “**PLASMA**” Collection and Aliquot Tube Label on the lavender-top EDTA tube. Place pre-printed “**PLASMA**” Collection and Aliquot Tube Labels on the (21) 0.5 ml siliconized cryovial tubes with clear caps and lavender stickers. Place pre-printed “**BUFFY COAT**” Collection and Aliquot Tube Label on the (1) 2 ml cryovial with a blue lid.
3. Please ensure that aliquots are kept in numerical order (by specimen number) throughout the aliquoting and shipping process.
4. Using a blood collection set and a holder, collect blood into the **EDTA (Lavender-Top) Blood Collection Tube (10 ml)** using your institution's recommended procedure for standard venipuncture technique.

The following techniques shall be used to prevent possible backflow:

- a. Place donor's arm in a downward position.
 - b. Hold tube in a vertical position, below the donor's arm during blood collection.
 - c. Release tourniquet as soon as blood starts to flow into tube.
 - d. Make sure tube additives do not touch stopper or end of the needle during venipuncture.
5. Allow at least 10 seconds for a complete blood draw to take place in each tube. **Ensure that the blood has stopped flowing into the tube before removing the tube from the holder.** The tube with its vacuum is designed to draw 10 ml of blood into the tube.
 - a. If complications arise during the blood draw, please note the difficulties on the ‘Biological Sample and Shipment Notification Form’. Do not attempt to draw an additional EDTA tube at this time. Process blood obtained in existing EDTA tube.
 6. **CRITICAL STEP: Immediately after blood collection, gently invert/mix (180 degree turns) the EDTA tube 5 times.**

7. **CRITICAL STEP: Immediately after inverting the EDTA tube, place it on wet ice until centrifugation begins.**
 - Preferably within 30 minutes of blood collection, centrifuge balanced tubes for 10 minutes at 2000 RCF (x g) at 4°C. **It is critical that the tubes be centrifuged at the appropriate speed and temperature to ensure proper plasma separation (see worksheet in [Appendix A](#) to calculate RPM.** Equivalent rpm for spin at 2000 x g
 - While centrifuging, remember to record all times, temperatures and spin rates on the Biological Sample and Shipment Notification Form.
 - Plasma samples need to be spun, aliquoted, and placed in the freezer within 2 hours from the time of collection.
 - Record time aliquoted on the Biological Sample Shipment and Notification Form.

8. Remove the plasma, being careful not to agitate the packed red blood cells at the bottom of the tube. Tilt the tube and place a disposable pipette tip along the lower side of the wall without touching the pellet (buffy coat) so that plasma is not contaminated (see below). Transfer plasma into the pre-labeled cryovials with the lavender stickers on the caps. Aliquot 0.25 ml per cryovial (total vials = 16-21 with 0.25 ml each). The EDTA tube should yield, on average, 5 ml of plasma for a total of 16-21 0.5 ml siliconized cryovial tubes per subject with 0.25 ml per cryovial tube. Be sure to only place **plasma** in cryovials with lavender stickers and labeled with “PLASMA” labels. Take caution not to disturb the red blood cells at the bottom of the tube. If there is extra plasma left, use 1 extra cryovial provided for another <0.25 ml aliquot of plasma. **If a residual aliquot (<0.25 ml) is created, document the sample number and volume on the Biological Sample and Shipment Notification Form.**



NOTE: When pipetting plasma from the plasma tube into the cryovials, be very careful to pipette the plasma top layer only, leaving the buffy coat and the red blood cell layers untouched.

Plasma Aliquots
(16 to 21 possible)

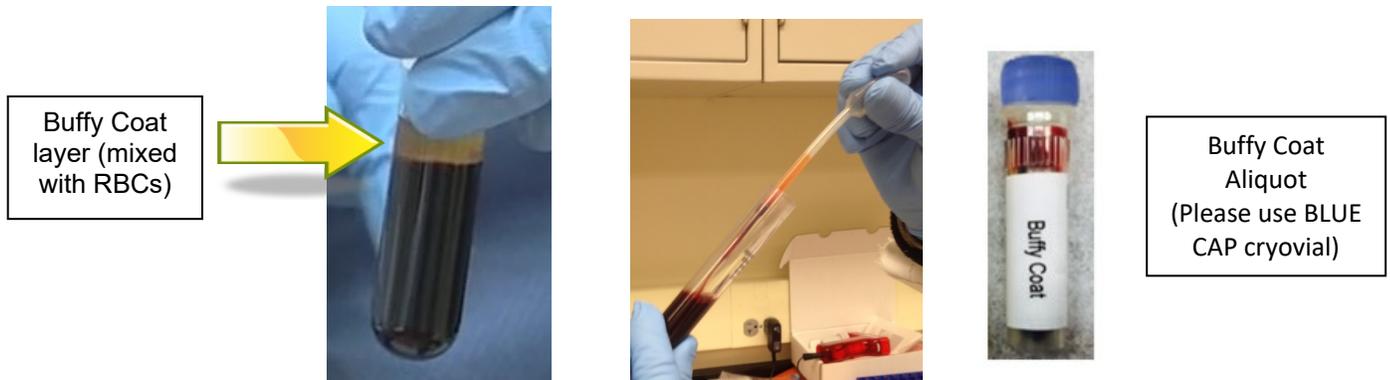


Close up view of 0.5 ml
Siliconized Cryovial



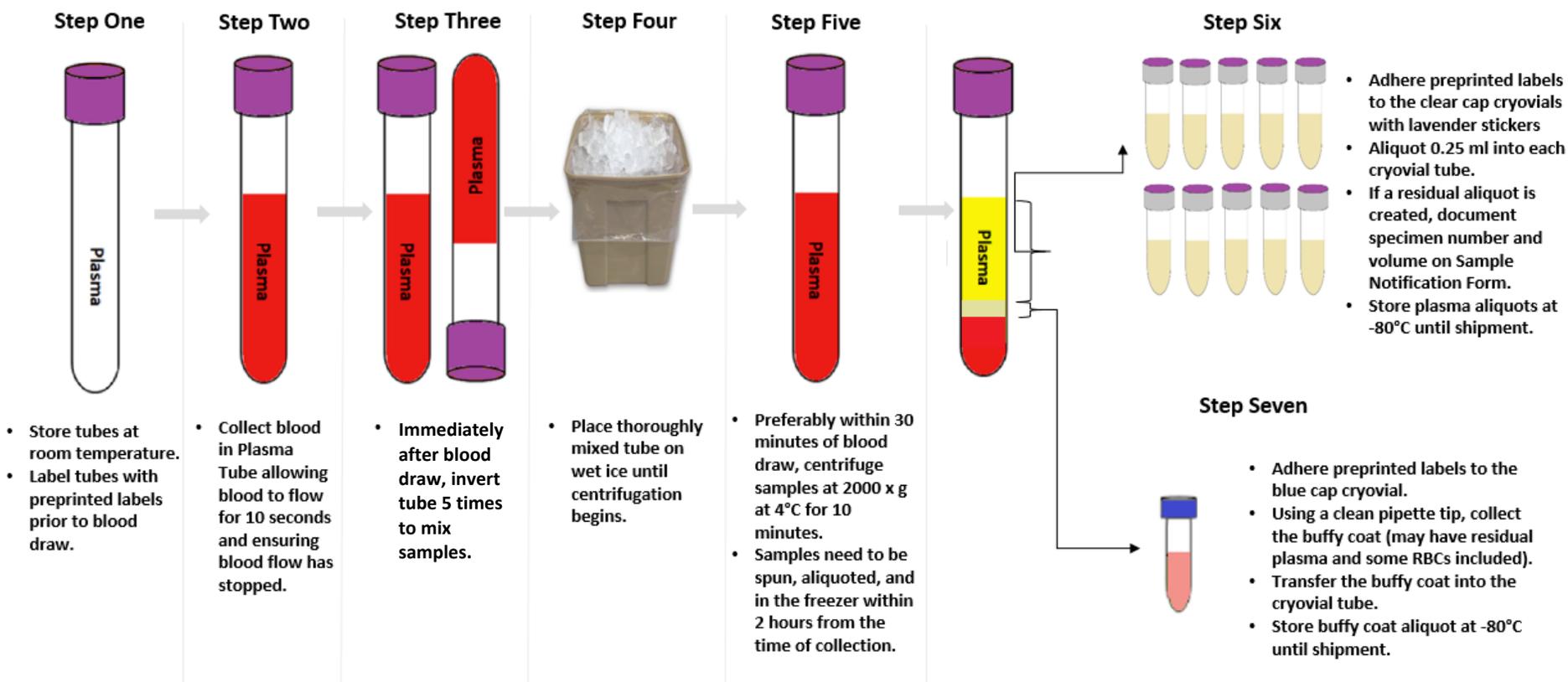
9. Place the labeled cryovials in the 25 cryovial box and place on dry ice. Transfer to **-80°C Freezer when possible**. Store all samples at **-80°C until shipped** to NCRAD on dry ice. Record time aliquots placed in freezer and storage temperature of freezer on Biological Sample Shipment and Notification Form.

10. After plasma has been removed from the EDTA (Lavender-Top) Blood Collection Tube (10 ml), aliquot buffy coat layer (in the top layer of cells, the buffy coat is mixed with RBCs-see figure) into labeled cryovial with blue cap using a micropipette. All of the buffy coat will be placed into one cryovial. The buffy coat aliquot is expected to have a reddish color from the RBCs. Be sure to place buffy coat into cryovial with the blue cap and “BUFFY COAT” label.



11. Dispose of tube with red blood cell pellet according to your site’s guidelines for disposing of biomedical waste.
12. Place the labeled cryovial in the 25 cryovial box and place on dry ice. Transfer to **-80°C Freezer when possible**. Store all samples at **-80°C until shipped** to NCRAD on dry ice.

Plasma and Buffy Coat Preparation (10ml Lavender-Top Tube)



6.6 Sodium Heparin (Green-Top) Blood Collection Tube (10 ml) for Karyotyping

Important Note

The Green-Top Sodium Heparin tubes are for the sites to use to obtain blood for karyotyping for full or partial trisomy 21 by the local clinical lab. This tube is drawn at baseline only. Sites should check with their local lab for the amount of blood needed and for the storage and transport conditions.

Draw blood in the following order:

1. Serum Separator (Gold-Top) Tube for Serum x 2 (*DS Participants only*)
2. EDTA (Lavender-Top) Tube for DNA and Plasma
3. Sodium Heparin (Green-Top) Tube for Karyotyping (*Baseline Visit Only; DS Participants Only*)

7.0 INCOMPLETE OR DIFFICULT BLOOD DRAWS

Important Note

If challenges arise during the blood draw process, it is advised that the phlebotomist discontinue the draw. Attempt to process and submit any blood-based specimens that have already been collected to NCRAD.

Redraws will not be scheduled for samples submitted to NCRAD as participants are seen longitudinally.

Situations may arise that prevent study coordinators from obtaining the total amount scheduled for biospecimens. In these situations, please follow the below steps:

1. *If the biospecimens at a scheduled visit are partially collected:*
 - a. Attempt to process and submit any samples that were able to be collected during the visit
 - b. Document difficulties on the 'Biological Sample and Shipment Notification Form' prior to submission to NCRAD
 - i. Indicate blood draw difficulties at the bottom of the 'Biological Sample and Shipment Notification Form' within the "Notes" section.
 - ii. Complete the 'Biological Sample and Shipment Notification Form' with tube volume approximations and number of aliquots created.

- c. Contact a NCRAD coordinator and alert them of the challenging blood draw
2. *If the biospecimens at a scheduled visit **are not** collected:*
- a. Contact the NiAD Global Coordinator and a NCRAD coordinator to alert them of the challenging blood draw or circumstances as to why biospecimens were not collected.
 - b. Schedule participant for a longitudinal visit.
 - i. If baseline samples were unable to be drawn, please draw the Sodium Heparin (Green-Top) Tube for Karyotyping during the next visit.

8.0 PACKAGING AND SHIPPING INSTRUCTIONS

ALL study personnel responsible for shipping should be certified in biospecimen shipping. If not available at your University, please contact NCRAD with questions and information regarding resources.

| Sample Type | NiAD | Processing/ Aliquoting | Tubes to NCRAD | Ship |
|---|------|---|-------------------|--------|
| Whole blood (Gold-Top SST) for isolation of serum Drawn for DS Participants ONLY | Yes | 0.25 ml serum aliquots per 0.5 ml siliconized cryovial (clear caps with RED stickers) | 16-21 | Frozen |
| Whole blood (Lavender-Top EDTA) for isolation of plasma & buffy coat (for DNA extraction) Drawn for DS Participants and Sibling Controls | Yes | 0.25 ml plasma aliquots per 0.5 ml siliconized cryovial (clear caps with LAVENDER stickers) | 16-21 | Frozen |
| | Yes | 1 ml buffy coat aliquot per 2.0 ml cryovial (BLUE cap) | 1 | Frozen |

IMPORTANT!
**FROZEN SAMPLES MUST BE SHIPPED
MONDAY-WEDNESDAY ONLY!**

Specimens being shipped to NCRAD should be considered as Category B UN3373 specimens and as such must be triple packaged and compliant with IATA Packing Instructions 650. *See the Latest Edition of the IATA Regulations for complete documentation.*

***** Packing and Labeling Guidelines *****

- The primary receptacle (frozen cryovials) must be leak proof and must not contain more than 1L total.
- The secondary packaging (biohazard bag) must be leak proof and if multiple blood tubes are placed in a single secondary packaging, they must be either individually wrapped or separated to prevent direct contact with adjacent blood tubes.
- Absorbent material must be placed between the primary receptacle (within the cryovial box containing the frozen cryovials) and the secondary packaging. The absorbent material should be of sufficient quantity in order to absorb the entire contents of the specimens being shipped. Examples of absorbent material are paper towels, absorbent pads, cotton balls, or cellulose wadding.
- A shipping manifest of specimens being shipped must be included between the secondary and outer packaging.
- The outer shipping container must display the
- following labels:
 - ✓ Sender's name and address
 - ✓ Recipient's name and address
 - ✓ Responsible Person
 - ✓ The words "Biological Substance, Category B"
 - ✓ UN3373
 - ✓ Class 9 label including UN 1845, and net weight of dry ice contained



Triple packaging consists of a primary receptacle(s), a secondary packaging, and a rigid outer packaging. The primary receptacles must be packed in secondary packaging in such a way that, under normal conditions of transport, they cannot break, be punctured, or leak their contents into the secondary packaging. Secondary packaging must be secured in outer packaging with suitable cushioning material. Any leakage of the contents must not compromise the integrity of the cushioning material or of the outer packaging.

8.1 Frozen Shipment Instructions

1. Contact FedEx to confirm service is available and schedule package to be picked up.
2. Notify NCRAD of shipment by emailing NCRAD coordinators at: alzstudy@iu.edu
Attach the following to the email:
 - Completed Biological Sample and Shipment Notification Form to the email notification.
(See [Appendix B](#) for an example of the NCRAD sample form)
 - If email is unavailable please call NCRAD and do not ship until you've contacted and notified NCRAD coordinators about the shipment in advance.
3. Place all frozen labeled aliquots of plasma and buffy coat in the cryovial cryobox.
 - i. Each cryobox holds 25 samples and there will be approximately 40 cryovial samples for DS Participants. Please place plasma and buffy coat aliquots within one cryobox and the serum aliquots within the second cryobox (for DS Participants). (21 plasma, 21 serum, and 1 buffy coat) per DS Participant blood draw (see below). Sibling Controls will have ONE cryobox with plasma and buffy coat aliquots only.



One cryobox to contain
Serum Aliquots.



One cryobox to contain Plasma
and Buffy Coat Aliquots.

- ii. Cryoboxes should contain all of the specimens from the same patient, per time point.
 - iii. **Batch shipping should be performed every 3 months or when specimens from 5 participants accumulates, whichever is sooner.**
4. Label the outside of the cryoboxes with the kit number label. Place Plasma/Buffy Coat aliquots within one cryobox and the serum aliquots in another cryobox. For DS Participants, BOTH cryoboxes should be placed within the SAME biohazard bag. For Sibling Controls, ONE cryobox should be placed within the biohazard bag. These biohazard bags are large enough to contain one or two cryoboxes.
5. Place the cryoboxes in the clear plastic biohazard bag (do NOT remove the absorbent material found in the bag) and seal according to the instructions on the bag.



Place kit
number label on
each cryobox

Plasma, Buffy Coat, and Serum Cryoboxes for cases placed in clear biohazard bag

6. Place approximately 2-3 inches of dry ice in the bottom of the Styrofoam shipping container.
7. Place the biohazard bag into the provided Styrofoam-lined shipping container on top of the dry ice. Please ensure that cryoboxes are placed so the cryovials are upright in the shipping container (as pictured below).
8. Fully cover the cryoboxes with approximately 2 inches of dry ice.
9. The inner Styrofoam shipping container must contain approximately 30-45 lbs (or 21kg) of dry ice. The dry ice should entirely fill the inner box to ensure the frozen state of the specimens.

Full Shipping Container with
Batched Samples and Dry Ice



10. Replace the lid on the Styrofoam carton. Place the completed Biological Sample and Shipment Notification Form in the package on top of the Styrofoam lid for each patient specimen, and close and seal the outer cardboard shipping carton with packing tape.

11. Complete the FedEx return airbill with the following information:
 - a. Section 1, "From": fill in your name, address, phone number, and Site FedEx Account Number.
 - b. Section 2, "Your Internal Billing Reference": add any additional information required by your site.
 - c. Section 6, "Special Handling and Delivery Signature Options": under "Does this shipment contain dangerous goods?" check the boxes for "Yes, Shipper's Declaration not required" and "Dry Ice". Enter the number of packages (1) x the net weight of dry ice in kg.
 - d. Section 7, "Payment", check sender and bill transportation costs to your site's study FedEx account number.



12. Complete the Class 9 UN 1845 Dry Ice label (black and white diamond) with the following information:

- a. Your name and return address
- b. Net weight of dry ice in kg (must match amount on the airbill)
- c. Consignee name and address:

NCRAD
IU School of Medicine
351 West 10th Street
TK-217
Indianapolis, IN 46202

- d. Do not cover any part of this label with other stickers, including pre-printed address labels.

13. Apply all provided warning labels and the completed FedEx return airbill to the outside of package, taking care not to overlap labels.

| |
|--|
| IMPORTANT! |
| Complete the required fields on the FedEx return airbill and Class 9 Dry Ice label, or FedEx may reject or return your package. |

14. Hold packaged samples in -80°C freezer until time of FedEx pick-up/drop-off.

15. Specimens should be sent to the below address via FedEx Priority Overnight. Frozen shipments should be sent Monday through Wednesday to avoid shipping delays on Thursday or Friday. FedEx does not replenish dry ice if shipments are delayed or held over during the weekend.

NCRAD
351 West 10th Street
TK-217
Indianapolis, IN 46202
Phone: 1-800-526-2839

16. Use FedEx tracking to ensure the delivery occurs as scheduled and is received by NCRAD. Please notify NCRAD by email (alzstudy@iu.edu) that a shipment has been sent and include the FedEx tracking number in your email.

*****Important Note*****

For frozen shipments, include no more than ten cryovial boxes (separated by patient within 5 biohazard bags) per shipping container in order to have room for a sufficient amount of dry ice to keep samples frozen up to 24 hours.

The labeled, processed, aliquoted, and frozen cryovials of serum, plasma, and buffy coat will be shipped to NCRAD as outlined above.

**SHIP ALL FROZEN SAMPLES MONDAY - WEDNESDAY ONLY!
BE AWARE OF HOLIDAYS!!
BE AWARE OF INCIPIENT INCLEMENT WEATHER THAT MAY DELAY
SHIPMENT/DELIVERY OF SAMPLES**

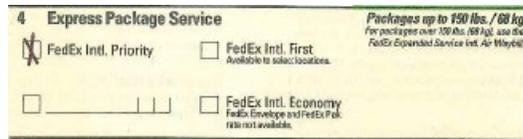
Remember to complete the Biological Sample and Shipment Notification ([Appendix B](#)), include a copy in your shipment **AND** notify the NCRAD Study Coordinator by email at alzstudy@iu.edu (include Fed Ex tracking number in email) **IN ADVANCE** to confirm the shipment.

In addition to tracking and reconciliation of samples, the condition and amount of samples received are tracked by NCRAD for each sample type. Investigators and clinical coordinators for each project are responsible to ensure the requested amounts of each fluid are collected to the best of their ability and that samples are packed with sufficient amounts of dry ice to avoid thawing in the shipment process.

8.2 International Shipping

1. All international shipments will utilize the same packing requirements as specified in [Section 8.1](#) (Frozen Shipping Instructions).
2. Two components are necessary for international shipments:
 1. International FedEx return airbill
 2. International Commercial Invoice
3. NCRAD will provide an International FedEx return airbill to all International sites. However, these international sites are welcome to utilize the FedEx electronic system.
 - a. Be sure to complete the FedEx return airbill with the following information:

- i. Section 1, From: Enter the date and your name, phone number, complete address, and FedEx account number.
- ii. Section 2, To: This information will be preprinted with NCRAD’s return address and phone number.
- iii. Section 3, Shipment information: This information does NOT replace a Commercial Invoice that is required for these shipments. Total Packages, Weight, and box dimensions are required. Be consistent between this International FedEx return airbill and the International Commercial Invoice.
 1. Do not declare the value of the shipment to be over \$2,500. This would require additional paperwork (a Shipper’s Export Declaration form).
- iv. Section 4, Express Package Services: Please check FedEx Intl. Priority for both Frozen and Ambient Shipments. (Pictured)



- v. Section 5, Packaging: Please select “Other” for Frozen Shipments and “FedEx Pak” for Ambient Shipments.
 - vi. Section 6, Special Handling: Please leave blank.
 - vii. Section 7 and 8, Payment: Check Sender and bill transportation costs to your site’s study FedEx account number. Duties and Taxes will also be billed to the sender. If your site requests information to be included as reference, please complete Section 8.
 - viii. Section 9, Required Signature: This section must be signed by the sender or department representative.
- b. International Commercial Invoice (See [Appendix C](#))
- i. The International Commercial Invoice must be completed and placed with the International return airbill.
 1. Include **ONE** original and **THREE** copies of this completed form with the FedEx return airbill.



- ii. Complete “Shipped From” with your name, address, and any additional contact information.
- iii. Complete “Shipped To, Consignee” with the NCRAD shipping address:

NCRAD
 IU School of Medicine
 351 West 10th Street
 TK-217
 Indianapolis, IN 46202
 Phone: 1-800-526-2839

- iv. Complete Number of Packages and Shipping weight to match the information recorded within the International FedEx return airbill.
- v. Immediately below the shipping weight is a section asking for the Country of Origin, Description of Goods, Quantity, Unit Price, and Total Price. Please be as detailed as possible within this section (example pictured below).

| COUNTRY OF ORIGIN & PROVINCE, IF CANADA PAIS D'ORIGINE ET PROVINCE, SI CANADA | DESCRIPTION OF GOODS DESCRIPTION DES MARCHANDISES | QUANTITY QUANTITE | UNIT PRICE PRIX UNITAIRE | TOTAL PRICE PRIX TOTAL |
|--|--|----------------------|-----------------------------|---------------------------|
| Canada, Vancouver | Non-Infectious, non-contagious, human Plasma and Buffy Coat sample | 1 Box (11 Aliquots) | 100.00 | 100.00 |

- vi. Tally the Total Price within the last column for all goods included in shipment and record appropriately.
 1. Reminder: the total price/value of the shipment should not exceed \$2,500.
- vii. Complete the final section with your signature.
- viii. Specimens should be sent to the below address via FedEx Priority Overnight. Ambient FedEx shipments should be sent Monday through Thursday. Frozen FedEx Shipments should only be sent Monday through Wednesday.
- ix. Use FedEx tracking to ensure the delivery occurs as scheduled and is received by NCRAD.

9.0 DATA QUERIES AND SAMPLE RECONCILIATION

The Laboratory worksheets must be completed on the day that samples are collected since they capture information related to the details of the sample collection and processing. These forms include information that will be used to reconcile sample collection and receipt, as well as information essential to future analyses.

The Alzheimer's Therapeutic Research Institute (ATRI) data collection team will be collaborating with NCRAD to reconcile information captured in the database compared to samples received and logged at NCRAD. Information that appears incorrect in the ATRI database will be queried through the standard system. Additional discrepancies that may be unrelated to data entry will be resolved with the Principal Investigator in a separate follow up communication. If applicable, a non-conformance report will be provided to sites on a monthly basis.

Data queries or discrepancies with samples shipped and received at NCRAD may result from:

- Missing samples
- Incorrect samples collected and shipped
- Damaged or incorrectly prepared samples
- Unlabeled samples, samples labeled with incomplete information, or mislabeled samples
- Discrepant information documented on the Biological Sample and Shipment Notification Form and logged at NCRAD compared to information entered into the ATRI database.
- Samples that are frozen and stored longer than one quarter at the site
- Use of an incorrect Biological or CSF Sample and Shipment Notification Form



10.0 APPENDICES LIST

[Appendix A: Rate of Centrifugation Worksheet](#)

[Appendix B: Biological Sample and Shipment Notification Form](#)

[Appendix C: International Commercial Invoice](#)



Appendix A Rate of Centrifuge Worksheet

Please complete and return this form by fax or email to the NCRAD Project Manager if you have any questions regarding sample processing. The correct RPM will be sent back to you.

Submitter Information

Name:

Site:

Submitter e-mail:

Centrifuge Information

Please answer the following questions about your centrifuge.

Centrifuge Type

Fixed Angle Rotor:

Swing Bucket Rotor:

Radius of Rotation (mm):

Determine the centrifuge's radius of rotation (in mm) by measuring distance from the center of the centrifuge spindle to the bottom of the device when inserted into the rotor (if measuring a swing bucket rotor, measure to the middle of the bucket).

Calculating RPM from G-Force:

$$RCF = \left(\frac{RPM}{1,000} \right)^2 \times r \times 1.118 \Rightarrow RPM = \sqrt{\frac{RCF}{r \times 1.118}} \times 1,000$$

RCF = Relative Centrifugal Force (G-Force)

RPM = Rotational Speed (revolutions per minute)

R = Centrifugal radius in mm = distance from the center of the turning axis to the bottom of centrifuge

Comments:

Please send this form to NCRAD Study Coordinator

317-321-2003 (Fax)

alzstudy@iu.edu



Appendix B



Appendix B



Participant ID: NIAD

Site ID:

Date: / /

Visit Mo:

Biological Sample and Shipment Notification Form

Please email or fax the form on or prior to the date of shipment.

| | | | |
|--|--|--|--|
| To: Kelley Faber Email: alzstudy@iu.edu FAX: 317-321-2003 Phone: 1-800-526-2839 | | | |
| <i>General Information:</i> | | | |
| From: _____ | | Date: _____ | |
| Phone: _____ | | Email: _____ | |
| Study: NIAD <input type="checkbox"/> DS Participant <input type="checkbox"/> Sibling Control - [Plasma/Buffy Coat (lavender top) only] | Kit #: _____ | | |
| Sex: <input type="checkbox"/> M <input type="checkbox"/> F Year of Birth: _____ | KIT BARCODE | | |
| FedEx tracking #: _____ | | | |
| <i>Blood Collection:</i> | | | |
| 1. Date Drawn: _____ [YYYYMMDD] | | 2. Time of Draw (24 hour clock): _____ [HHMM] | |
| 3. Last time subject ate (Date): _____ [YYYYMMDD] | | 4. Last time subject ate (24 hour clock): _____ [HHMM] | |
| 5. Was the EDTA tube placed on ice immediately after inverting tube 5 times until centrifugation began? <input type="checkbox"/> Yes <input type="checkbox"/> No | | | |
| <i>Blood Processing:</i> | | | |
| Plasma (EDTA/Lavender Top Tube) | | Serum (Serum Separator/Gold Top Tube) | |
| • Time spin started (24 hour clock): _____ [HHMM] | • Time spin started (24 hour clock) (30 minutes after draw time): _____ [HHMM] | | |
| • Duration of centrifuge: _____ [minutes] | • Duration of centrifuge: _____ [minutes] | | |
| • Temp of centrifuge: _____ °C • Rate of centrifuge: _____ x g | • Temp of centrifuge: _____ °C • Rate of centrifuge: _____ x g | | |
| Original volume drawn (1x10 mL EDTA tube): _____ mL | Original volume drawn (2x5 mL Serum tube): _____ mL | | |
| • Time aliquoted: _____ [HHMM] | • Time aliquoted: _____ [HHMM] | | |
| Number of 0.25 mL plasma aliquots created (16-20 total) (Siliconized cryovial): _____ x 0.25 mL | Number of 0.25 mL serum aliquots created (16-20 total) (Siliconized cryovial): _____ x 0.25 mL | | |
| • If applicable, volume of residual plasma aliquot (less than 0.25 mL) (Siliconized cryovial): _____ mL | • If applicable, volume of residual serum aliquot (less than 0.25 mL) (Siliconized cryovial): _____ mL | | |
| If applicable, specimen number of residual aliquot (Last four digits): _____ | If applicable, specimen number of residual aliquot (Last four digits): _____ | | |
| • Time aliquots placed in freezer (24 hour clock): _____ [HHMM] | • Time aliquots placed in freezer (24 hour clock): _____ [HHMM] | | |
| • Storage temperature of freezer: _____ °C | • Storage temperature of freezer: _____ °C | | |
| Buffy coat aliquot created (one per EDTA tube) (Blue cap cryovial): _____ mL | <i>Note: Bulleted items not entered into eCRF.</i> | | |
| Notes: _____ | | | |

Version 03.2020



Appendix C

International Commercial Invoice

FOR CUSTOMS CLEARANCE BY / POUR DÉDOUANEMENT PAR



INVOICE
FACTURE

STAPLE LINE RELEASE LABEL HERE

| | | | | |
|--|--|--|--|--|
| EXPORTER, SELLER, CONSIGNOR EXPORTATEUR, VENDEUR, EXPÉDITEUR | | TELEPHONE TÉLÉPHONE | SHIPPED FROM (IF DIFFERENT THAN EXPORTER) EXPÉDÉ (SI DIFFÉRENT DE L'EXPORTATEUR) | |
| U.S. IRS NUMBER OR S/S NUMBER | | REFERENCE RÉFÉRENCE | | |
| BUYER (IF SOLD) ACHÉTEUR (SI VENDU) | | TELEPHONE TÉLÉPHONE | SHIPPED TO, CONSIGNEE (IF NOT SOLD OR IF DIFFERENT THAN BUYER) EXPÉDÉ AU DESTINATAIRE (SI NON VENDU OU SI AUTRE QUE L'ACHÉTEUR) | |
| U.S. IRS NUMBER OR S/S NUMBER | | REFERENCE RÉFÉRENCE | U.S. IRS NUMBER OR S/S NUMBER | |
| PARTIES TO THIS TRANSACTION ARE LES TRANSACTIONNAIRES SONT | | COUNTRY OF FINAL DEST. (IF OTHER THAN U.S.A.) PAYS DE DESTINATION FINALE (SI AUTRE QUE LES E.-U.) | INVOICE NUMBER NUMÉRO DE FACTURE | |
| <input type="checkbox"/> RELATED ASSOCIÉS <input type="checkbox"/> NOT RELATED NON ASSOCIÉS | | | INVOICE DATE DATE DE LA FACTURE | |
| U.S. DUTY AND/OR BROKERAGE FORT / DROITS DE DOUANE AMÉRICAIN ET/OU FRAIS DE COURTOISAGE POUR | | | IMPORTING CARRIER/TRANSPORTEUR À L'IMPORTATION | |
| <input type="checkbox"/> EXPORTER (INCLUDED) (COMPRIS) OR FOR THE ACCOUNT OF OU POUR LE COMPTE DE | | <input type="checkbox"/> EXPORTER (NOT INCLUDED) (NON COMPRIS) | <input type="checkbox"/> BUYER ACHÉTEUR | <input type="checkbox"/> CONSIGNEE DESTINATAIRE |
| | | | DATE OF SALE DATE DE LA VENTE | |
| | | | DISCOUNTS ESCOMPTES | |
| | | | CURRENCY OF VALUE VALEUR EN DEVISES | |
| | | | <input type="checkbox"/> US | <input type="checkbox"/> CANADIAN CANADIENNES |
| | | | OTHER AUTRE | |
| MARKS AND NUMBERS MARIQUES ET NUMÉROS | | NUMBER AND KIND OF PACKAGES NOMBRE ET CATÉGORIE DE COLIS | SHIPPING WEIGHT POIDS DE L'EXPÉDITION | FREIGHT AMOUNT INCLUDED MONTANT DU FRET COMPRIS |
| | | | | FREIGHT AMOUNT TO BORDER MONTANT DU FRET À LA FRONTIÈRE |
| COUNTRY OF ORIGIN & PROVINCE IF CANADA PAYS D'ORIGINE ET PROVINCE, SI CANADA | DESCRIPTION OF GOODS DESCRIPTION DES MARCHANDISES | | QUANTITY QUANTITÉ | UNIT PRICE PRIX UNITAIRE |
| | | | | TOTAL PRICE PRIX TOTAL |
| Cargo Insurance is available through FedEx Trade Networks. Please contact Customer Service at 1.800.249.2953 for a competitive quote. L'assurance cargo est offerte par l'entremise de FedEx Trade Networks. Veuillez contacter le service à la clientèle au 1.800.249.2953 pour une estimation compétitive. | | | INVOICE TOTAL TOTAL DE LA FACTURE | |
| DECLARATION BY FOREIGN SHIPPER (COMPLETE IF GOODS ARE OF U.S. ORIGIN AND VALUE EXCEEDS \$2000.00) | | | | |
| I, _____, DECLARE THAT TO THE BEST OF MY KNOWLEDGE AND BELIEF THE ARTICLES HEREIN SPECIFIED WERE EXPORTED FROM THE UNITED STATES, FROM THE PORT OF _____ ON OR ABOUT _____ AND THAT THEY ARE RETURNED WITHOUT HAVING BEEN ADVANCED IN VALUE OR IMPROVED IN CONDITION BY ANY PROCESS OF MANUFACTURE OR OTHER MEANS. | | | | |
| SIGNATURE | | CAPACITY | NAME OF RESPONSIBLE EMPLOYEE OF EXPORTER NOM DE L'EMPLOYÉ RESPONSABLE CHEZ L'EXPORTATEUR | |
| To the best of the knowledge and belief of the preparer this invoice is true and complete and discloses the true prices, values, quantities, rebates, drawbacks, fees, commissions, royalties and any goods or services provided to the seller either free or at a reduced cost. | | PREPARER (IF OTHER THAN EXPORTER) PRÉPARATEUR (SI AUTRE QUE L'EXPORTATEUR) | | |

FORM 12 3/05 WV

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