ADCFB Protocol Update: V03.2026

Section	Change
Document Footer	Updated to "Version (3.2026)"
1.0	Added Biomarker Assay Lab.
3.3	Updated link to holiday closure page of NCRAD website: https://ncrad.org/contact/holiday-closures
4.0	Noted to contact NIABRICSOperations@mail.nih.gov to troubleshoot GUID issues.
5.2.1	Updated link to recommend consent language on NCRAD website: https://www.ncrad.org/bank-samples/sample-management/recommended-consent-language
5.2.2	Updated Plasma and Buffy Coat Aliquot Volumes in Blood Collection Table to reflect replacement of 1 x 1.5 ml plasma aliquot with 1 x 0.75 ml plasma aliquot and an anticipated buffy coat aliquot volume of 0.5 - 1.0 ml.
6.1	Updated Blood Collection Kit and Supplemental Supply Kits to reduce Purple Cap Cryovial counts and add Yellow Cap Cryovials. Updated Blood and CSF Collection kit contents to match supply order in Kit Request Module. Added supply list for ADCFB Blood Kit 30 – Michigan, ADCFB Blood Supplemental Supply Kit 10, and NCRAD Batch Frozen Shipping Kit. Added Yellow Cap Cryovials to Individual Supplies list.
6.3	• **IMPORTANT**: Each blood kit will now include a 2 ml yellow capped cryovial designated as the BAL (Biomarker Assay Lab) tube. This cryovial should be filled with 0.75 ml of plasma and is required for rapid returns of biomarker assay results. • Updated instructions, example diagram for filling plasma cryovials, and Cap Color table to note addition of yellow capped cryovial for BAL aliquot. Clarified expectations for aliquoting and recording sample barcodes and volumes. • Emphasized that empty cryovials should NOT be returned to NCRAD. • Updated table to include yellow capped cryovial and example photos of cap colors.
7.0	Updated Collection and Aliquot tube label examples to reflect changes to Sample Number formats.
7.2	Replaced references to Blood Sample and Shipment Notification Form with ADCFB Specimen Collection And Processing Form.

7.3	Specified labeling procedure for yellow capped BAL plasma aliquot. Updated existing cryobox example photos and added a 10 ml Collection example. Replaced references to Blood Sample and Shipment Notification Form with ADCFB Specimen Collection And Processing Form.
8.0	Updated maximum ADCFB collection volume to 15-20 ml. Replaced references to the CSF Sample and Shipment Notification Form with ADCFB Specimen Collection And Processing Form.
9.0	**IMPORTANT**: Added section 9.1 Shipping Notifications providing instructions on how to utilize the NCRAD Specimen Shipment Notification Form.
9.2	Ambient Packaging Instructions updated to section 9.2.
9.3	Frozen Packaging Instructions updated to section 9.3.
9.4	Ambient and Frozen Shipping Instructions updated to section 9.4.
9.2.1.2	Updated shipment notification instructions to use NCRAD Specimen Shipment Notification Form
9.2.1.7	Clarified expectations for paperwork to be included with package.
9.3.1.1	Updated shipment notification instructions to use NCRAD Specimen Shipment Notification Form
9.3.1.5	Emphasized packing limits for frozen shippers.
9.3.1.9	Clarified expectations for paperwork to be included with package.
10.0	**IMPORTANT**: Added Section 10.0 ADCFB Specimen Collection And Processing Form providing instructions for use of the required online sample collection and processing form.
11.0	Data Reconciliation updated to section 11.0.
12.0	Appendices updated to section 12.0.
Appendix C	Blood Sample and Shipment Notification Form: Added sections under Blood Processing for Volume and Specimen number of BAL plasma aliquot. Updated shipping notification instructions.
Appendix D	Updated shipping notification instructions.

Alzheimer's Disease Center Fluid Biomarkers

in collaboration with the

National Centralized Repository for Alzheimer's Disease and Related Dementias

Biospecimen Collection, Processing, and Shipment Manual of Procedures

Version 03.2026

TABLE OF CONTENTS

Contents

1.0 Abbreviations

AD	Alzheimer’s Disease
ADCFB	Alzheimer’s Disease Center Fluid Biomarkers
BAL	Biomarker Assay Lab
CSF	Cerebrospinal Fluid
DNA	Deoxyribonucleic Acid
EDTA	Ethylene Diamine Tetra-acetic Acid
GUID	Globally Unique Identifier
IATA	International Air Transport Association
NACC	National Alzheimer's Coordinating Center
NCRAD	National Centralized Repository for Alzheimer’s Disease and Related Dementias
PHI	Protected Health Information
RBC	Red Blood Cells
RCF	Relative Centrifugal Force
RPM	Revolutions Per Minute

2.0 Purpose

The collection of biofluids is an important part of the Alzheimer's Disease Center Fluid Biomarkers (ADCFB) Study. 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 biological samples for ADCFB study visits. It includes instructions for biofluid submission to NCRAD located in Indianapolis at Indiana University.

Centers may collect and send the following samples to NCRAD:

- PBMC
- Plasma
- Buffy Coat (DNA Extraction)
- Cerebrospinal Fluid (CSF)

This manual includes instructions for collection of blood and CSF, 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 specimens provided to NCRAD for the ADCFB protocol.

3.0 NCRAD Information

3.1 NCRAD Contacts

Tatiana Foroud, PhD, Core Leader
Phone: 317-274-2218

Kelley Faber, MS, CCRC, Senior Project Manager
Phone: 317-274-7360
Email: kelfaber@iu.edu

Stephanie Steidel, MS, Clinical Research Coordinator
Phone: 317-274-1685
Email: ssteidel@iu.edu

General NCRAD Contact Information
Phone: 1-800-526-2839 or 317-278-8413
Email: alzstudy@iu.edu
Website: www.ncrad.org

Sample Shipment Mailing Address
ADCFB at NCRAD
Indiana University School of Medicine
351 W. 10th St. TK-217
Indianapolis, IN 46202
Phone: 1-800-526-2839
Alternate phone: 1-317-278-8413

3.2 NCRAD 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.**

For packing and shipment details of samples, please refer to Section 9.0 of this protocol.

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

3.3 NCRAD Holiday Observations

Date	Holiday
January 1st	New Year's Day
3rd Monday in January	Martin Luther King, Jr Day
4th Monday in May	Memorial Day
June 19	Juneteenth
July 4	Independence Day
1st Monday in September	Labor Day
4th Thursday in November	Thanksgiving
4th Friday in November	Friday after Thanksgiving
December 25th	Christmas Day
December 26th – 31st	Winter Break

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 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 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/contact/holiday-closures for additional information.**

- 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.
- **Weekend/holiday delivery must be arranged in advance with NCRAD staff.**

4.0 Globally Unique Identifier (GUID)

The GUID is a participant ID that allows researchers to share data specific to a study participant, without exposing personally identifiable information. A GUID is made up of random alpha-numeric characters and does not include any PHI in the identifier. By using GUIDs in your research data, the system can associate a single research participant's genetic, imaging, and clinical assessment data even if the data was collected at different locations or throughout different studies. No PHI will be sent to NCRAD, only the GUID.

Please contact NIABRICSOperations@mail.nih.gov to troubleshoot any login or usage issues.

To create a GUID follow these steps:

1. Create an account: https://bricsguid.nia.nih.gov/portal/jsp/login.jsp
2. Once you have an account, go to the GUID Tool – Create GUID
3. To open the 'Launch GUID Tool' you will need to have Java installed on your device
4. In order to generate a GUID, the following PHI is required (Appendix A):
    - Complete legal given (first) name of participant at birth
    - If the participant has a middle name
    - Complete legal family (last) name of participant at birth
    - Day of birth
    - Month of birth
    - Year of birth
    - Name of city/municipality in which participant was born
    - Country of birth

5.0 ADCFB Laboratory Collection

5.1 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 and hub
- Microcentrifuge tube rack
- Sharps bin and lid
- Wet Ice Bucket
- Wet ice
- Pelleted dry ice

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 x g with refrigeration to 4°C
- -80°C Freezer

In order to ship specimens, you must provide:

- Pelleted dry ice (approximately 15 pounds for small batches and 45 pounds for large batches)

5.2 Biospecimens Sent to NCRAD

Samples are to be submitted according to the shipping methods outlined in Section 9.0. Guidelines for the processing, storage location, and timing of sample collection are listed in the tables below.

5.2.1 Biofluid Collection Schedule

Depending on the approved blood volume, centers will collect and send the following blood-based biospecimens to NCRAD:

Blood-based Biospecimen Collection Table

	Total Blood Volume Collected			
Biospecimen	**20 ml**	**30 ml**	**40 ml**	**50 ml**
Plasma	X	X	X	X
Buffy Coat (DNA)	X	X	X	X
PBMC			X	X

Whole blood is collected in up to three purple-top EDTA tubes and up to two green-top sodium heparin (NaHep) tubes for shipment to NCRAD. If collected, the sodium heparin tubes are shipped to NCRAD on the day of the participant visit (Monday through Thursday only). The 10 ml EDTA tubes are processed locally into plasma and buffy coat fractions; they are then aliquoted, frozen at the study site, and shipped to NCRAD.

If desired, sites may also collect and send CSF samples at each annual visit:

CSF Biospecimen Collection Table

Biospecimen	Annual visits
CSF	X

Consent forms must specify that any biological samples and de-identified clinical data may be shared with academic and/or industry collaborators through NCRAD. Recommended consent language can be found on the NCRAD website at: https://www.ncrad.org/bank-samples/sample-management/recommended-consent-language. A copy of the consent form for each participant should be kept on file by the site investigator.

5.2.2 Biofluid Collection Charts

Blood Collection Table

Draw Order*	Collection Tube	Drawn At	Specimen Type	Aliquot Volume	Total Number of Aliquots	Shipping Temperature
1**	**2 Sodium Heparin (Green-Top) Blood Collection Tubes (10 ml)**	Visit 1	Whole Blood	N/A	N/A	Ambient
2	**2 or 3 EDTA (Purple-Top) Blood Collection Tubes (10 ml)**	Each visit	Plasma	1.5 ml plasma aliquots	Up to 9	Frozen
		Each visit	Plasma	0.75 ml plasma aliquot	1	Frozen
		Each visit	Buffy Coat	0.5 - 1.0 ml buffy coat aliquots	Up to 3	Frozen

**Chart shows full 50ml blood collection. Combination of tubes being drawn may vary but always need to follow this order for the tubes that are drawn.*
***If drawing blood for PBMCs, Sodium Heparin tubes **must** be the first tubes into which blood is collected. If not, the EDTA tubes will be first.*

CSF Collection Table

Collection Tube	Drawn At	Specimen Type	Aliquot Volume	Total Number of Aliquots	Shipping Temperature
Sterile Container	Each visit	CSF	1.5 ml CSF aliquots	Up to 14	Frozen

6.0 Specimen Collection Kits, Shipping Kits, and Supplies

NCRAD will provide: 1) Blood sample collection kits for research specimens to be stored at NCRAD, the Blood Supplemental Supply Kit, the Frozen Shipment Kit; 2) CSF collection kits including Lumbar Puncture (LP) trays, the CSF Supplemental Supply Kit; and 3) clinical lab supplies (with the exception of pelleted dry ice and equipment supplies listed in Section 5.1). The provided materials include blood tubes, pipettes, LP trays (when applicable), boxes for plasma, buffy coat, and CSF aliquots, as well shipping materials to send biospecimens to NCRAD. Kit number labels, site and PTID labels, and collection tube and aliquot labels will all be provided by NCRAD. Details regarding the blood kits are found in this Manual of Procedures. Collection tube and aliquot labels will be preprinted 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 7.1.

Depending on the allowed blood volume, Centers may order one of four types of kits:

ADCFB Kits

	Total Blood Volume Collected			
	20 ml	**30 ml**	**40 ml**	**50 ml**
Kit Type	20	30	40	50

6.1 NCRAD Specimen Collection Kit Contents

Collection kits contain the following (for each participant) and provide the necessary supplies to collect samples from a given participant. 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.*

Blood Collection Kits

ADCFB Blood Kit 20

Quantity	ADCFB Blood Kit 20 Components
1	Large Plastic Bag
2	EDTA (purple-top) blood collection tube (10 ml)
1	15 ml conical polypropylene tube (orange cap)
1	Cryovial box (holds up to 25 cryovials)
5	Cryovial (2.0 ml) with purple cap
1	Cryovial (2.0 ml) with yellow cap
1	Cryovial (2.0 ml) with blue cap
2	Cryovial (2.0 ml) with clear cap
4	Disposable graduated transfer pipettes (3 ml)
11	Preprinted Collection Tube and Aliquot Labels
3	Preprinted Kit Number Label
3	Label for handwritten Site and PTID

ADCFB Blood Kit 30

Quantity	ADCFB Blood Kit 30 Components
1	Large Plastic Bag
3	EDTA (purple-top) blood collection tube (10 ml)
1	50 ml conical polypropylene tube (blue cap)
1	Cryovial box (holds up to 25 cryovials)
8	Cryovial (2.0 ml) with purple cap
1	Cryovial (2.0 ml) with yellow cap
1	Cryovial (2.0 ml) with blue cap
3	Cryovial (2.0 ml) with clear cap
4	Disposable graduated transfer pipettes (3 ml)
16	Preprinted Collection Tube and Aliquot Labels
3	Preprinted Kit Number Label
4	Label for handwritten Site and PTID

ADCFB Blood Kit 40

Quantity	ADCFB Blood Kit 40 Components
1	Large Plastic Bag
2	Sodium Heparin (green-top) blood collection tube (10 ml)
2	EDTA (purple-top) blood collection tube (10 ml)
1	15 ml conical polypropylene tube (orange cap)
1	Cryovial box (holds up to 25 cryovials)
5	Cryovial (2.0 ml) with purple cap
1	Cryovial (2.0 ml) with yellow cap
1	Cryovial (2.0 ml) with blue cap
2	Cryovial (2.0 ml) with clear cap
4	Disposable graduated transfer pipettes (3 ml)
13	Preprinted Collection Tube and Aliquot Labels
3	Preprinted Kit Number Label
5	Label for handwritten Site and PTID

ADCFB Blood Kit 50

Quantity	ADCFB Blood Kit 50 Components
1	Large Plastic Bag
2	Sodium Heparin (green-top) blood collection tube (10 ml)
3	EDTA (purple-top) blood collection tube (10 ml)
1	50 ml conical polypropylene tube (blue cap)
1	Cryovial box (holds up to 25 cryovials)
8	Cryovial (2.0 ml) with purple cap
1	Cryovial (2.0 ml) with yellow cap
1	Cryovial (2.0 ml) with blue cap
3	Cryovial (2.0 ml) with clear cap
4	Disposable graduated transfer pipettes (3 ml)
18	Preprinted Collection Tube and Aliquot Labels
3	Preprinted Kit Number Label
6	Label for handwritten Site and PTID

Site Specific Blood Collection Kits

ADCFB Blood Kit 10

***Sites with restricted blood collection volumes ONLY**

Quantity	ADCFB Blood Kit 10 Components
1	Large Plastic Bag
1	EDTA (purple-top) blood collection tube (10 ml)
1	Cryovial box (holds up to 25 cryovials)
2	Cryovial (2.0 ml) with purple cap
1	Cryovial (2.0 ml) with yellow cap
1	Cryovial (2.0 ml) with blue cap
1	Cryovial (2.0 ml) with clear cap
2	Disposable graduated transfer pipettes (3 ml)
6	Preprinted Collection Tube and Aliquot Labels
3	Preprinted Kit Number Label
2	Label for handwritten Site and PTID

ADCFB Blood Kit 20 - Vanderbilt

***Vanderbilt ONLY**

Quantity	ADCFB Vanderbilt Blood Kit 20 Components
1	Large Plastic Bag
1	EDTA (purple-top) blood collection tube (10 ml)
1	Sodium Heparin (green-top) blood collection tube (10 ml)
1	Cryovial box (holds up to 25 cryovials)
2	Cryovial (2.0 ml) with purple cap
1	Cryovial (2.0 ml) with yellow cap
1	Cryovial (2.0 ml) with blue cap
1	Cryovial (2.0 ml) with clear cap
2	Disposable graduated transfer pipettes (3 ml)
7	Preprinted Collection Tube and Aliquot Labels
3	Preprinted Kit Number Label
3	Label for handwritten Site and PTID

ADCFB Blood Kit 30 – UAB*

***University of Alabama Birmingham ONLY**

Quantity	ADCFB Blood Kit 30 - UAB Components
1	Large Plastic Bag
2	EDTA (purple-top) blood collection tube (10 ml)
1	Sodium Heparin (green-top) blood collection tube (10 ml)
1	15 ml conical polypropylene tube (orange cap)
1	Cryovial box (holds up to 25 cryovials)
5	Cryovial (2.0 ml) with purple cap
1	Cryovial (2.0 ml) with yellow cap
1	Cryovial (2.0 ml) with blue cap
2	Cryovial (2.0 ml) with clear cap
4	Disposable graduated transfer pipettes (3 ml)
12	Preprinted Collection Tube and Aliquot Labels
3	Preprinted Kit Number Label
4	Label for handwritten Site and PTID

ADCFB Blood Kit 30 – Michigan*

***University of Michigan ONLY**

Quantity	ADCFB Blood Kit 30 - UAB Components
1	Large Plastic Bag
1	EDTA (purple-top) blood collection tube (10 ml)
2	Sodium Heparin (green-top) blood collection tube (10 ml)
1	Cryovial box (holds up to 25 cryovials)
2	Cryovial (2.0 ml) with purple cap
1	Cryovial (2.0 ml) with yellow cap
1	Cryovial (2.0 ml) with blue cap
1	Cryovial (2.0 ml) with clear cap
2	Disposable graduated transfer pipettes (3 ml)
8	Preprinted Collection Tube and Aliquot Labels
3	Preprinted Kit Number Label
4	Label for handwritten Site and PTID

CSF Collection Kits

ADCFB LP Kits*

**Sites must specify 22- or 24-gauge kit when ordering from NCRAD.*

Quantity	LP Kit Components
1	Sprotte needle, 22 or 24 gauge X 3.5” (90mm)
1	Introducer needle, 1 mm x 30 mm
1	Hypodermic needle, 22 gauge x 1.5”
1	Plastic syringe, (3 ml, luer lock) with 25G x 5/8” needle attached
4	Polypropylene syringe (5 ml, luer lock)
1	Needle stick pad
1	Adhesive bandage
1	Drape, fenestrated, 2 tabs, paper, 18” x 26”
2	Towel, 13.5” x 18”
6	Gauze pad, 2” x 2”
3	Sponge stick applicator
2	Lidocaine 1%, 5 ml
1	Povidone-Iodine Topical Solution, 0.75 oz

ADCFB CSF Kits

Quantity	CSF Kit Components
1	Large plastic bag
3	15 ml conical polypropylene tube-individually wrapped
1	50 ml conical polypropylene tube-individually wrapped
1	Cryovial box (holds up to 25 cryovials)
13	Cryovial tube (2.0 ml) with orange cap
1	Cryovial tube (2.0 ml) with yellow cap
1	Cryovial tube (2.0 ml) with blue cap
14	Pre-printed Collection Tube and Aliquot Label
3	Pre-printed Kit Number label
3	Label for handwritten Patient ID

ADCFB 1.5 ml CSF Kits

Quantity	CSF Kit Components
1	Small resealable plastic bag
1	Cryovial tube (2.0 ml) with orange cap
1	Pre-printed Collection Tube and Aliquot Label
3	Pre-printed Kit Number label

ADCFB Supplemental Supply Kits

Limit 1 Supplemental supply kit per site per year.

ADCFB Blood Supplemental Supply Kit 10

Quantity	ADCFB Blood Supplemental Supply Kit 20 Components
1	Large Plastic Bag
5	EDTA (purple-top) blood collection tube (10 ml)
1	Cryovial box (holds up to 25 cryovials)
10	Cryovial (2.0 ml) with purple cap
5	Cryovial (2.0 ml) with yellow cap
5	Cryovial (2.0 ml) with blue cap
5	Cryovial (2.0 ml) with clear cap
5	Disposable graduated transfer pipettes (3 ml)
10	Label for handwritten Site and PTID

ADCFB Blood Supplemental Supply Kit 20

Quantity	ADCFB Blood Supplemental Supply Kit 20 Components
1	Large Plastic Bag
10	EDTA (purple-top) blood collection tube (10 ml)
5	15 ml conical polypropylene tube (orange cap)
5	Cryovial box (holds up to 25 cryovials)
25	Cryovial (2.0 ml) with purple cap
5	Cryovial (2.0 ml) with yellow cap
5	Cryovial (2.0 ml) with blue cap
10	Cryovial (2.0 ml) with clear cap
10	Disposable graduated transfer pipettes (3 ml)
15	Label for handwritten Site and PTID

ADCFB Blood Supplemental Supply Kit 30

Quantity	ADCFB Blood Supplemental Supply Kit 20 Components
1	Large Plastic Bag
15	EDTA (purple-top) blood collection tube (10 ml)
5	50 ml conical polypropylene tube (blue cap)
5	Cryovial box (holds up to 25 cryovials)
40	Cryovial (2.0 ml) with purple cap
5	Cryovial (2.0 ml) with yellow cap
5	Cryovial (2.0 ml) with blue cap
15	Cryovial (2.0 ml) with clear cap
10	Disposable graduated transfer pipettes (3 ml)
15	Label for handwritten Site and PTID

ADCFB Blood Supplemental Supply Kit 40

Quantity	ADCFB Blood Supplemental Supply Kit 20 Components
1	Large Plastic Bag
10	EDTA (purple-top) blood collection tube (10 ml)
10	Sodium Heparin (green-top) blood collection tube (10 ml)
5	15 ml conical polypropylene tube (orange cap)
5	Cryovial box (holds up to 25 cryovials)
25	Cryovial (2.0 ml) with purple cap
5	Cryovial (2.0 ml) with yellow cap
5	Cryovial (2.0 ml) with blue cap
10	Cryovial (2.0 ml) with clear cap
10	Disposable graduated transfer pipettes (3 ml)
15	Label for handwritten Site and PTID

ADCFB Blood Supplemental Supply Kit 50

Quantity	ADCFB Blood Supplemental Supply Kit 20 Components
1	Large Plastic Bag
10	EDTA (purple-top) blood collection tube (10 ml)
15	Sodium Heparin (green-top) blood collection tube (10 ml)
5	50 ml conical polypropylene tube (blue cap)
5	Cryovial box (holds up to 25 cryovials)
40	Cryovial (2.0 ml) with purple cap
5	Cryovial (2.0 ml) with yellow cap
5	Cryovial (2.0 ml) with blue cap
15	Cryovial (2.0 ml) with clear cap
10	Disposable graduated transfer pipettes (3 ml)
15	Label for handwritten Site and PTID

CSF Supplemental Supply Kit

Quantity	CSF Supplemental Supply Kit Components
10	50 ml conical polypropylene tube-individually wrapped (blue cap)
20	15 ml conical polypropylene tube-individually wrapped (blue cap)
20	Cryovial tube (2.0 ml) with orange cap
5	Cryovial tube (2.0 ml) with blue cap
5	Cryovial tube (2.0 ml) with yellow cap
3	Dry Ice Shipping Label and UN3373 Label
10	Small biohazard bags with absorbent sheet
5	3 ½" × 22 Sprotte needle with Introducer (90mm)
10	Adhesive Spot Bandage

ADCFB Shipping Kits

NCRAD Ambient Shipping Kit

Quantity	NCRAD Ambient Shipping Kit Components
1	Plastic biohazard bag with absorbent sheet
1	Small IATA shipping box with insulated cooler
1	Small refrigerant pack
1	UN3373 Biological Substance Category B label
1	List of contents card
1	UPS Airbill Sleeve
1	UPS Laboratory Pak

NCRAD Frozen Shipping Supply Kit (Small Shippers)

Quantity	Frozen Shipping Kit Components for Blood-Based Biomarkers
1	Large Plastic Bag
3	Plastic Biohazard bag with absorbent sheet (small)
1	UPS Airbill Sleeve
1	Shipping box/Styrofoam container (Small– Outer dimensions: 10x12x16”)
1	UN3373 label
1	UPS Dry ice shipping label

NCRAD Frozen Shipping Supply Kit (Large Shippers)

Quantity	Frozen Shipping Kit Components for Blood-Based Biomarkers
1	Large Plastic Bag
8	Plastic Biohazard bag with absorbent sheet (small)
1	UPS Airbill Sleeve
1	Shipping box/Styrofoam container (Large – Outer dimensions: 17x17x17”)
1	UN3373 label
1	UPS Dry ice shipping label

NCRAD Batch Frozen Shipping Kit (Large Shippers)

Quantity	Frozen Shipping Kit Components for Blood-Based Biomarkers
1	Large Plastic Bag
2	Plastic Biohazard bag with absorbent sheet (Large)
1	UPS Airbill Sleeve
1	Shipping box/Styrofoam container (Large – Outer dimensions: 17x17x17”)
1	UN3373 label
1	UPS Dry ice shipping label
4	81-cell Cardboard freezer boxes
8	Large rubber bands

Individual Supplies

Quantities	Items Available upon request within the NCRAD kit module
By Request	EDTA (Purple-Top) Blood Collection Tube (10 ml)
By Request	Sodium Heparin (green-top) blood collection tube (10 ml)
By Request	15 ml conical polypropylene tube (orange cap)
By Request	50 ml conical polypropylene tube (blue cap)
By Request	15 ml conical polypropylene tube(individually wrapped, blue cap)
By Request	50 ml conical polypropylene tube (individually wrapped, blue cap)
By Request	Cryovial box (holds up to 25 cryovials)
By Request	Cryovial (2.0 ml) with purple cap
By Request	Cryovial (2.0 ml) with yellow cap
By Request	Cryovial (2.0 ml) with blue cap
By Request	Cryovial (2.0 ml) with clear cap
By Request	Cryovial (2.0 ml) with orange cap
By Request	Disposable graduated transfer pipette (3 ml)
By Request	Site and PTID Labels
By Request	Small Shipping box/Styrofoam container
By Request	Large Shipping box/Styrofoam container
By Request	UPS Airbill Sleeve
By Request	UPS Clinic Pack
By Request	Small biohazard bag with absorbent sheet
By Request	Large biohazard bag with absorbent sheet
By Request	Warning label packet (UN3373 label and UPS dry ice shipping label)
By Request	UN3373 label
By Request	UPS dry ice shipping label
By Request	Fragile label
By Request	Arrow label
By Request	Bubble Wrap Tube Sleeve

6.2 Kit Supply to Study Sites

Each 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: http://kits.iu.edu/adcfb to request additional kits and follow the prompts to request the desired supplies.

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

6.3 Filling Cryovials (Plasma and CSF)

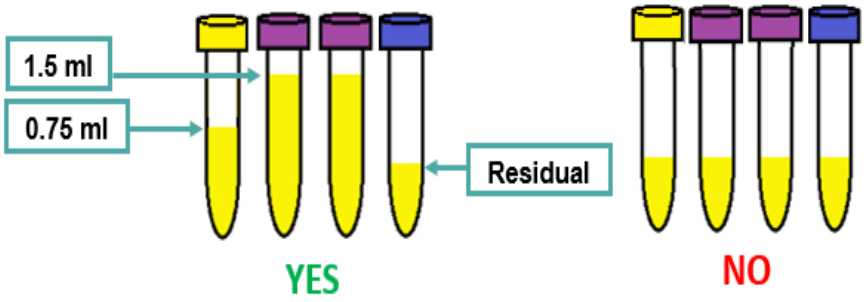
In order to ensure that NCRAD receives a sufficient amount of sample for processing and storage, and to avoid cracking of the tubes prior to shipment, each aliquot tube should be filled to the assigned volume after processing is completed (refer to detailed processing instructions for average yield per sample). Over-filled tubes may burst once placed in the freezer, resulting in a loss of sample.

The standard aliquot volume for ADCFB plasma and CSF is 1.5 ml (1500 ul). Standard plasma aliquots should be placed in purple capped tubes. Standard CSF aliquots should be placed in orange capped tubes.

In order to facilitate rapid return of plasma biomarker data, each blood collection kit includes one yellow capped cryovial. The yellow capped cryovial should be filled with a set volume of 0.75 ml (750 ul) and is designated as a BAL (Biomarker Assay Lab) aliquot. Please note that if the BAL aliquot is not received or is underfilled, biomarker assay results may be delayed several weeks.

Any material remaining after standard and BAL aliquots are created should be shipped to NCRAD as a residual volume aliquot placed in a blue capped cryovial. No residual volume is too low to return to NCRAD.

Ship *all* material to NCRAD. Fill as many aliquot tubes as possible. For example, if 2.7 ml of a plasma sample is obtained, fill 1 purple capped cryovial with 1.5 ml, fill 1 yellow capped cryovial with 0.75 ml, and one blue capped cryovial with the remaining residual volume (0.45 ml).



Please note: It is critical for the rapid return of biomarker data and for the integrity of future studies using these samples that study staff note if a plasma aliquot tube contains a nonstandard volume (below 1.5 ml).

Please highlight the 0.75 ml BAL plasma aliquot by utilizing the yellow capped cryovial provided in each kit. Please record the volume and either the last four digits or full barcode of the yellow cap cryovial on the ADCFB Specimen Collection and Processing Form.

Please highlight any residual aliquots by utilizing the blue capped cryovials provided in each kit. Please record the volume and either the last four digits or full barcodes of any residual aliquots on the ADCFB Specimen Collection and Processing Form.

If there are any unused cryovials, do not send the empty cryovials to NCRAD. Empty tubes received at NCRAD are assumed biohazardous and disposed of accordingly at the cost of the ADCFB study. These unused cryovials (ensure labels are removed and discarded) can be saved as local supplemental supplies or the cryovials can be disposed of per local requirements.

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

Example	Cap Color	Sample Type
	Yellow	Plasma (BAL aliquot – 0.75 ml)
	Purple	Plasma (1.5 ml)
	Clear	Buffy Coat
	Orange	CSF
	Blue	Residual sample (Plasma and/or CSF)

7.0 Blood Collection and Processing Procedures

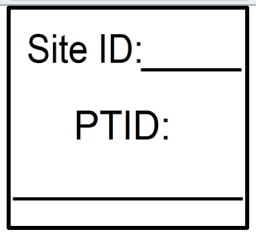
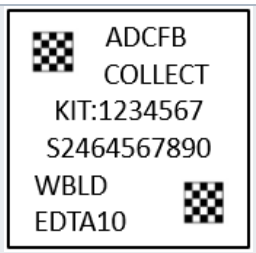
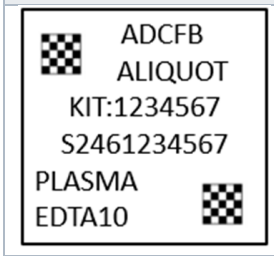
7.1 Labeling Samples

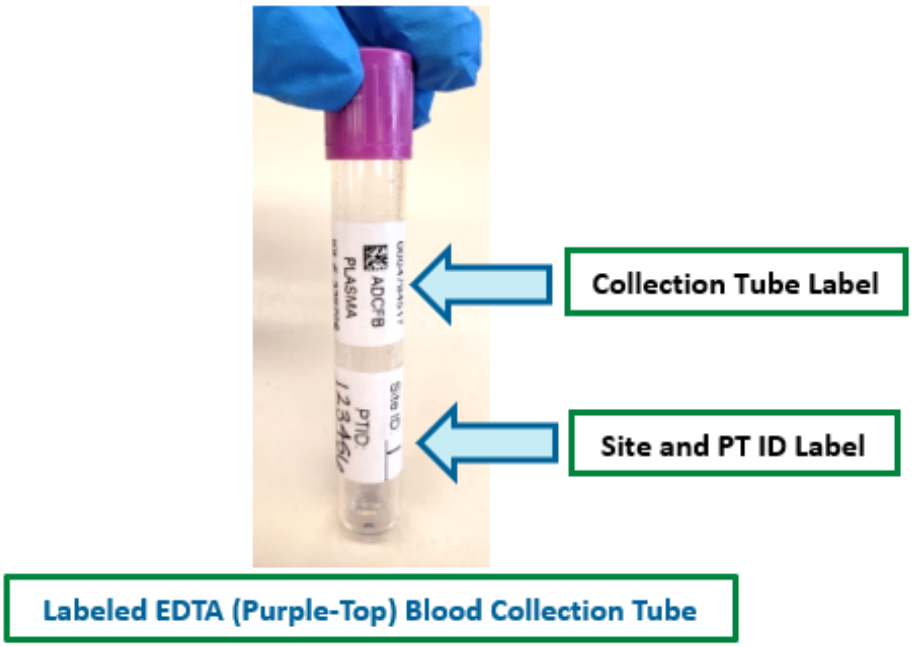
*****Important Note*****

In order to ensure the highest quality samples are collected, it is essential to follow the specific collection 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.

Label Type Summary

1. **Kit Number Label**
2. **Site and PTID Label**
3. **Collection Tube Labels**
4. **Aliquot Labels**

Example	Description
	**Kit Number Labels** tie together all specimens collected from one participant at one visit. They should be placed on each cryobox, and in the designated location on the Blood or CSF Sample and Shipment Notification Forms (if used).
	**Site and PTID Labels** are used to document the individual's unique Site and PTID. Place one label on each blood collection tube.
	**Collection Tube Labels** are placed on each collection tube. Labels are marked with COLLECT on the second line.
	**Aliquot Labels** are placed on each cryovial tube. Aliquot Labels are marked with ALIQUOT on the second line.



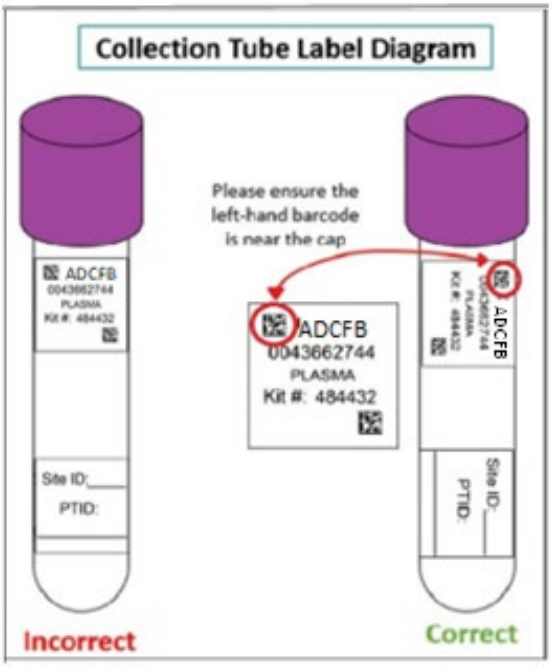
****Important Note****

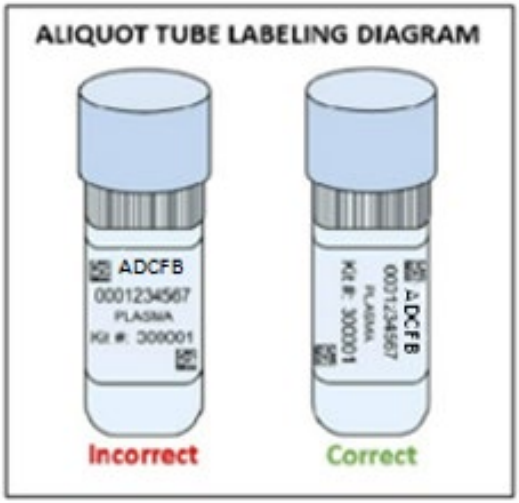
Each collection tube will have two labels: the collection tube label and the Site and PTID Label. Be sure to place labels in the same configuration consistently among tubes, with the barcoded label near the top of the tube and the handwritten Site and PTID label near the bottom of the tube.

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

- Place Collection Tube and Aliquot Labels on **ALL** collection tubes and cryovials **BEFORE** sample collection. This should help to ensure the label properly adheres to the tube before exposure to moisture or different temperatures.
- Using a fine point permanent marker, fill-in and place the Site and PTID Labels on the EDTA (purple-top) tubes **BEFORE** sample collection. These labels are placed on collection tubes in addition to the Collection Tube Label.
- The Collection Tube Labels contain a 2D barcode on the top left-hand side and bottom right-hand side of the label. Place the top left barcode toward the tube cap.
- Place label **horizontally** on the tube (wrapped around sideways if the tube is upright).

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.





7.2 Whole Blood Collection with 10 ml Sodium Heparin (Green-Top) Tube for PBMC

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

Once drawn, sodium heparin tubes MUST be shipped to NCRAD the day of collection via UPS Next Day Air service. This is to ensure the specimens have the most viable cells available at extraction.

These samples should only be collected Monday-Thursday. DO NOT collect these samples on Fridays.

1. Store empty sodium heparin tubes at room temperature, 64°F - 77°F (18 °C – 25 °C) before use.

2. Place completed site and PTID label and preprinted **PBMC** collection tube label on each of the sodium heparin (green-top) blood collection tubes. Place labels such that a clear window remains visible down the length of the tube as shown below to ensure buffy coat can be seen by NCRAD lab staff during processing.

3. Using a blood collection set and a holder, collect blood into the 10 ml sodium heparin tubes 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 last collection tube.
   d. Make sure tube additives do not touch the stopper or the end of the needle during venipuncture.

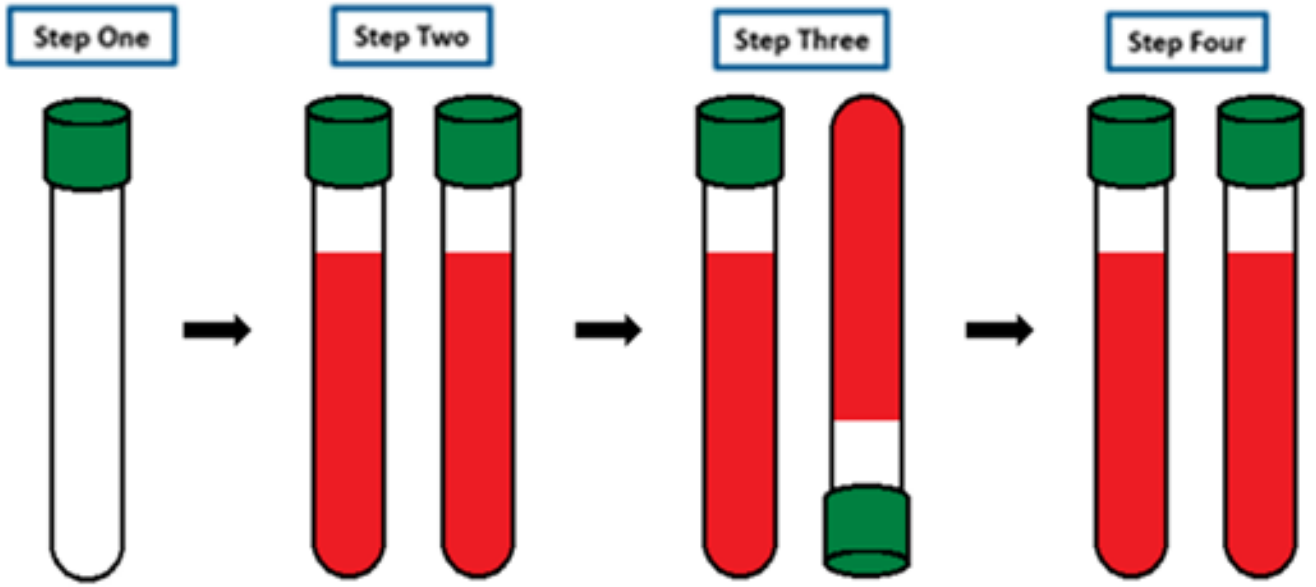
4. Allow at least 10 seconds for a complete blood draw to take place in the tube. **Ensure that the blood has stopped flowing into each tube before removing the tube from the holder.** The tube with its vacuum is designed to draw 10 ml of blood into the tube.

5. Immediately after blood collection, gently invert/mix (180-degree turns) each tube 8-10 times.

6. Ship the unprocessed sodium heparin (green-top) blood collection tubes **ambient** to NCRAD the day of the participant visit. Please see Section 9.1 for detailed ambient shipping instructions.

7. Complete ADCFB Specimen Collection And Processing Form.

PBMC Preparation (10 ml Sodium Heparin Tube)



- Store tubes at room temp.
- Label tubes with pre-printed PTID and collection tube labels prior to blood draw.

- Collect blood in Sodium Heparin tubes allowing blood to flow for 10 seconds, and ensuring blood flow has stopped.

- Immediately after blood draw, invert tubes 8-10 times to mix sample.

- Store tubes at room temp. until shipment.
- Ship ambient same day as blood draw.

Check expiration dates of tubes before collection to make sure tubes are not expired!

7.3 Whole Blood Collection with 10 ml EDTA (Purple-Top) Tube for Plasma and Buffy Coat

1. Store empty EDTA tubes at room temperature, 64°F - 77°F (18 °C – 25 °C) before use.

2. Set centrifuge to 4°C to pre-chill before use.

3. Place completed site and PTID Label and preprinted **PLASMA** Collection Tube Label on the purple-top EDTA tubes. Place preprinted **PLASMA** Aliquot Labels on the 2 ml cryovials with purple caps, the 2 ml cryovial with a yellow cap, and the 2 ml cryovial with a blue cap (if necessary, for residual). Place preprinted **BUFFY COAT** Aliquot Label on the 2 ml cryovials with clear caps.

4. Using a blood collection set and a holder, collect blood into the **EDTA (Purple-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 participant's arm in a downward position.
   b. Hold tube in a vertical position, below the participant’s arm during blood collection.
   c. Release tourniquet as soon as blood starts to flow into last collection 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 in the Comments section of the ADCFB Specimen Collection And Processing Form. Do not attempt to draw an additional EDTA tube at this time. Process blood obtained in existing EDTA tube.

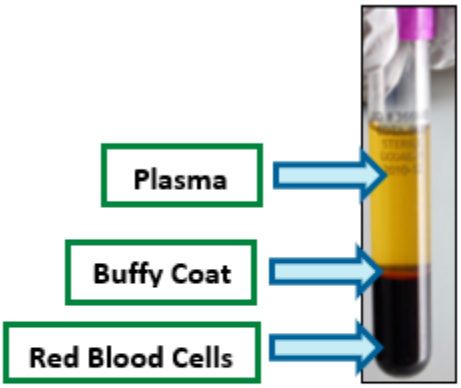
6. Immediately after blood collection, gently invert/mix (180 degree turns) the EDTA tube 8-10 times.

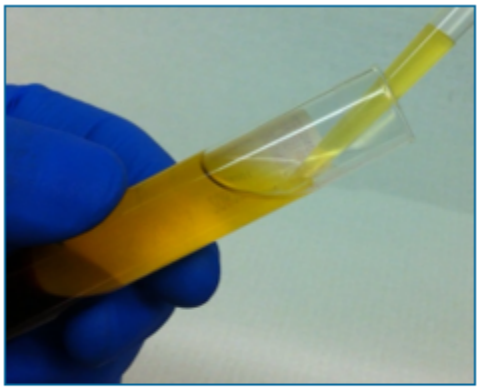
7. Immediately after inverting the EDTA tube, place it on wet ice until centrifugation begins.

8. Centrifuge balanced tubes for 10 minutes at 2000 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 B to calculate equivalent RPM for spin at 2000 x g).**
    a. While centrifuging, remember to record all times, temperatures and spin rates in the ADCFB Specimen Collection And Processing Form.
    b. Record original volume drawn for each tube in fields provided in the ADCFB Specimen Collection And Processing Form.
    c. Plasma samples need to be spun, aliquoted, and placed in the freezer within 2 hours from the time of collection.
    d. Record time aliquoted in the ADCFB Specimen Collection And Processing Form.

9. Remove the plasma by tilting the tube and placing the pipette tip along the lower side of the wall being careful not to agitate the packed red blood cells at the bottom of the collection tube.

10. Each EDTA tube should yield, on average, 4-5 ml of plasma. Transfer plasma from all EDTA tubes into the 15 ml conical tube (for 20 ml collections) or the 50 ml conical tube (for 30 ml collections) and gently invert 3 times. Aliquot 1.5 ml plasma per cryovial. Be sure to only place **plasma** in cryovials with purple and yellow caps and labeled with **PLASMA** labels**.** Place residual plasma (<1.5 ml) in the blue-capped cryovial. **If a residual aliquot (<1.5 ml) is created, document the specimen number and volume in the ADCFB Specimen Collection And Processing Form.**

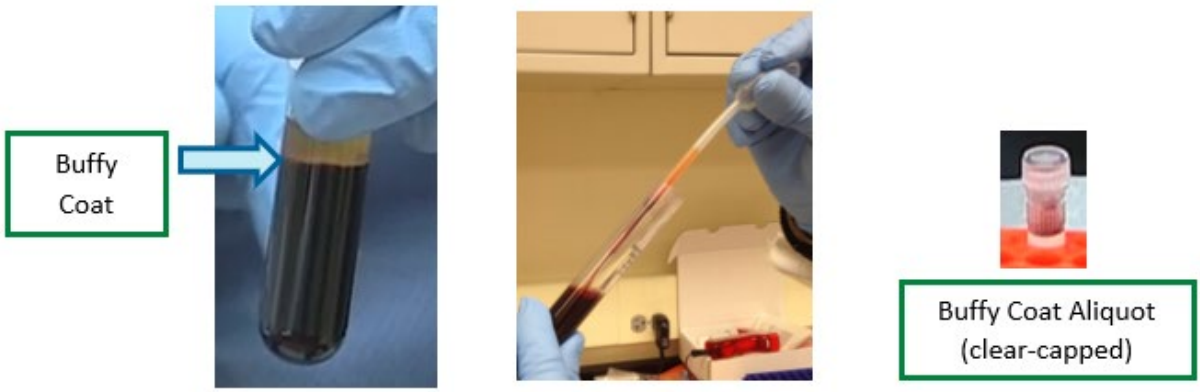




NOTE: When pipetting plasma from the EDTA tube into the 15 ml or 50 ml conical tube, be very careful to pipette the plasma top layer only, leaving the buffy coat and the red blood cell layers untouched.

11. Place the labeled cryovials in the 25 cell cryobox and place on pelleted dry ice. **Transfer to -80°C Freezer when possible**. Store all samples at **-80°C until shipped** to NCRAD on pelleted dry ice. Record time aliquots frozen and storage temperature of freezer in ADCFB Specimen Collection And Processing Form.

12. After plasma has been removed from the EDTA (Purple-Top) Blood Collection Tubes (10 ml), aliquot the buffy coat layer (in the top layer of cells, the buffy coat is mixed with RBCs-see figure) from one EDTA tube into a labeled, clear-capped cryovial using a micropipette. The buffy coat aliquot is expected to have a reddish color from the RBCs. Be sure to only place the buffy coat from one EDTA tube into each cryovial. Repeat this step for the second and third EDTA tubes (if collecting 30ml total), placing these buffy coats into the second and third clear-capped cryovials.



13. Dispose of collection tube with red blood cell pellet according to your site's guidelines for disposing of biomedical waste.

14. Record the specimen number and volumes of the EDTA tubes and corresponding buffy coat samples in the ADCFB Specimen Collection And Processing Form.

15. Place the labeled cryovials in the 25 cell cryobox and place on pelleted dry ice. **Transfer to -80°C Freezer when possible.** Store all samples at -80°C until shipped to NCRAD on pelleted dry ice. Record time aliquots frozen and storage temperature of freezer in the ADCFB Specimen Collection And Processing Form.

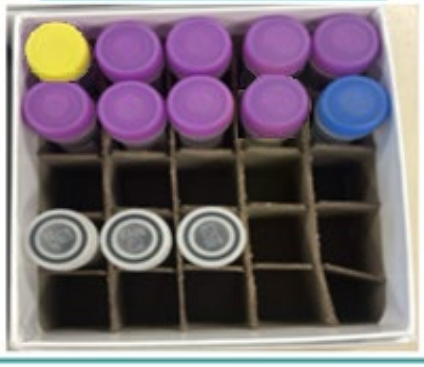
10 ml Collection

Plasma Aliquots (up to 4 possible) and Buffy Coat (1)

20 ml or 40 ml Kit

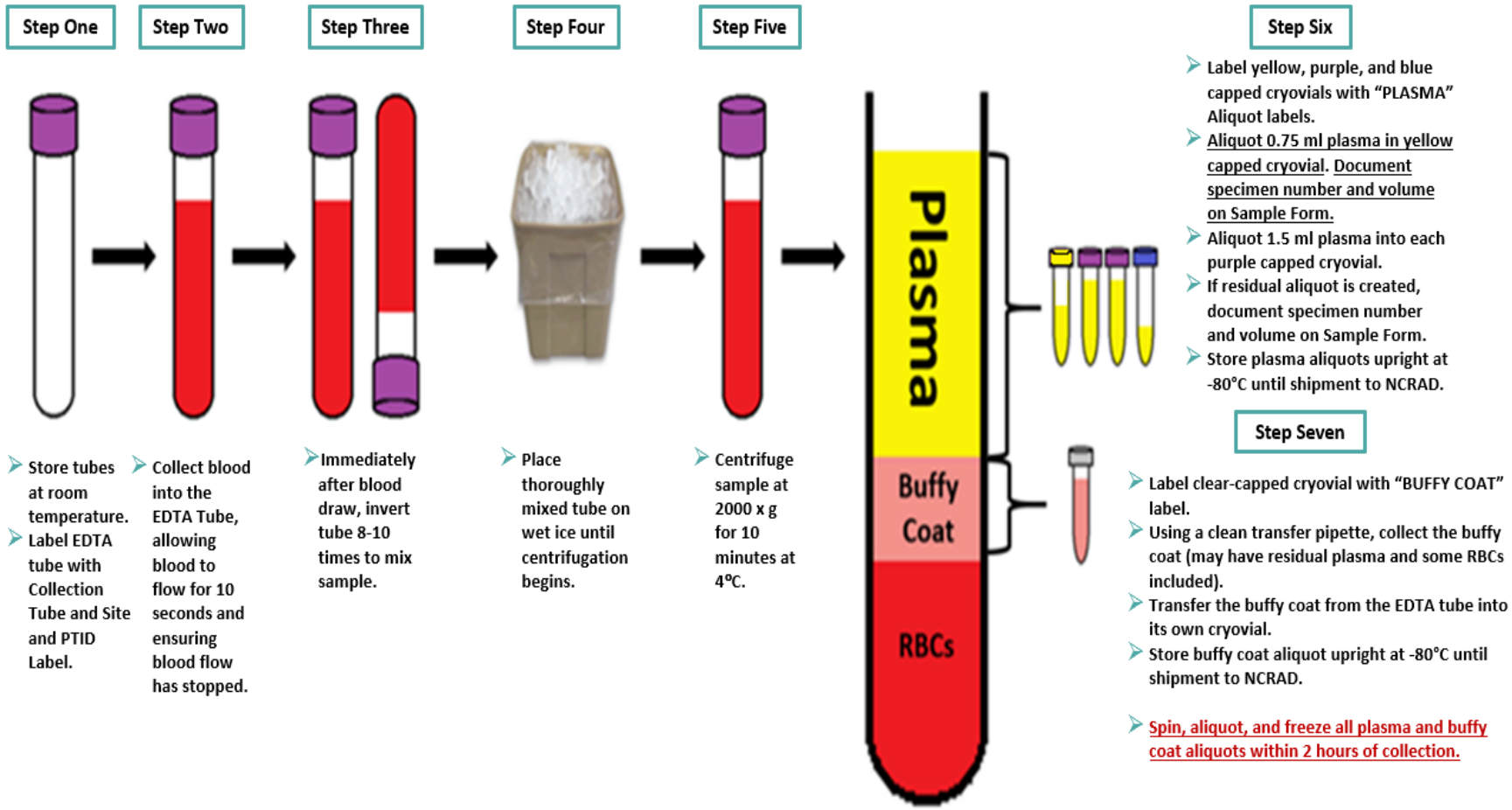
Plasma Aliquots (up to 7 possible) and Buffy Coats (2)

30 ml or 50 ml Kit



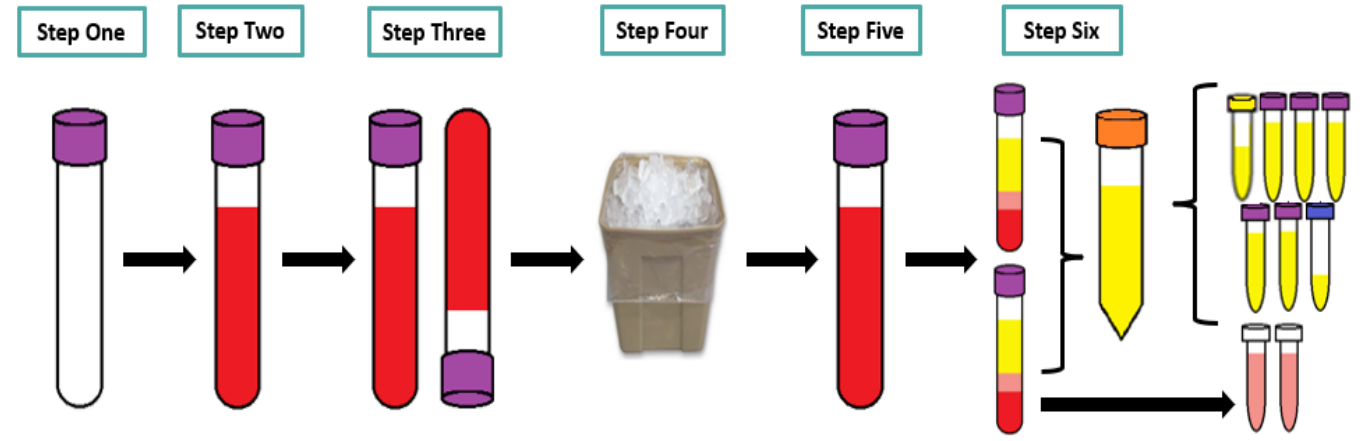
Plasma Aliquots (up to 10 possible) and Buffy Coats (3)

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



Check expiration dates of tubes before collection to make sure tubes are not expired!

Plasma and Buffy Coat Preparation EDTA Purple-Top Tube (2 x 10 ml)



Step One

- Store tubes at room temp.
- Each tube should be labeled with Collection Tube and Site and PTID Labels.

Step Two

- Collect blood into each EDTA Tube, allowing blood to flow for 10 seconds and ensuring blood flow has stopped.

Step Three

- Immediately after blood draw, invert tubes 8-10 times to mix samples.

Step Four

- Place thoroughly mixed tubes on wet ice until centrifugation begins.

Step Five

- Centrifuge samples at 2000 x g for 10 minutes at 4°C.

Step Six

- Pool all plasma from the 2 EDTA tubes into a 15 ml conical tube and invert gently 3 times to mix the plasma.

Step Seven

- Label yellow, purple, and blue capped cryovials with "PLASMA" Aliquot labels.
- Aliquot 0.75 ml plasma in yellow capped cryovial. Document specimen number and volume on Sample Form.
- Aliquot 1.5 ml plasma into each purple capped cryovial.
- If residual aliquot is created, document specimen number and volume on Sample Form.
- Store plasma aliquots upright at -80°C until shipment to NCRAD.

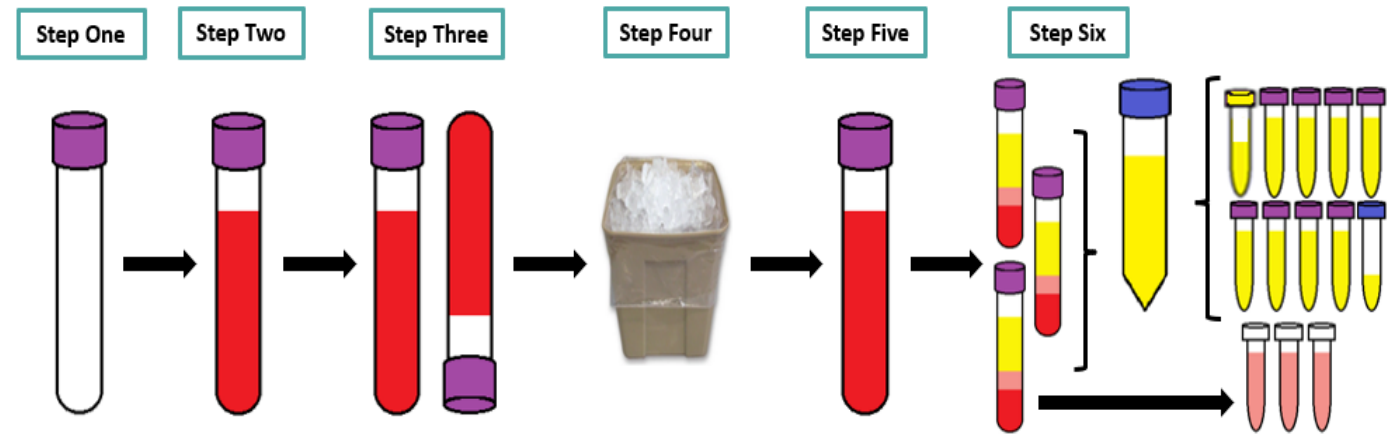
Step Eight

- Label clear-capped cryovials with "BUFFY COAT" labels.
- Using a clean transfer pipette, collect the buffy coat (may have residual plasma and some RBCs included).
- Transfer the buffy coat from each EDTA tube into its own cryovial.
- Store buffy coat aliquots upright at -80°C until shipment to NCRAD.

- Spin, aliquot, and freeze all plasma and buffy coat aliquots within 2 hours of collection.

Check expiration dates of tubes before collection to make sure tubes are not expired!

Plasma and Buffy Coat Preparation EDTA Purple-Top Tube (3 x 10 ml)



- Store tubes at room temp.
- Each tube should be labeled with Collection Tube and Site and PTID Labels.

- Collect blood into each EDTA Tube, allowing blood to flow for 10 seconds and ensuring blood flow has stopped.

- Immediately after blood draw, invert tubes 8-10 times to mix samples.

- Place thoroughly mixed tube on wet ice until centrifugation begins.

- Centrifuge samples at 2000 x g for 10 minutes at 4°C.

- Pool all plasma from the 3 EDTA tubes into a 50 ml conical tube and invert gently 3 times to mix the plasma.

- Label yellow, purple, and blue capped cryovials with "PLASMA" Aliquot labels.
- Aliquot 0.75 ml plasma in yellow capped cryovial. Document specimen number and volume on Sample Form.
- Aliquot 1.5 ml plasma into each purple capped cryovial.
- If residual aliquot is created, document specimen number and volume on Sample Form.
- Store plasma aliquots upright at -80°C until shipment to NCRAD.

Step Eight

- Label clear-capped cryovials with "BUFFY COAT" labels.
- Using a clean transfer pipette, collect the buffy coat (may have residual plasma and some RBCs included).
- Transfer the buffy coat from each EDTA tube into its own cryovial.
- Store buffy coat aliquots upright at -80°C until shipment to NCRAD.

- Spin, aliquot, and freeze all plasma and buffy coat aliquots within 2 hours of collection.

Check expiration dates of tubes before collection to make sure tubes are not expired!

8.0 Cerebrospinal Fluid Collection and Processing

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

CSF samples should be collected in the morning before breakfast and after an overnight fast. There should be a minimum 6-hour fast before collection of biomarker fluids and CSF. Only water is permitted until blood draws and the lumbar puncture are completed.

There are general guidelines to follow regarding CSF Collection.

- Begin by confirming participant consented to lumbar puncture (LP) before scheduling the procedure and again prior to performing procedure.
- If LP and PET scan are done on the same day, LP should be completed prior to the PET scan; otherwise, there should be at least 12 hours between LP and PET scan.
- LP should occur after, or a minimum of 72 hours prior, to an MRI scan.
- Do NOT use any extension tubing due to the tendency of manufactured plastic tubing to bind beta amyloid peptides and other important AD biomarkers.
- If LP was attempted but unsuccessful in obtaining CSF, a second attempt under fluoroscopy (if deemed appropriate by site clinician) is allowed.
- LP under fluoroscopy is permitted, if needed. Site personnel should advise the participant that use of fluoroscopy (x-rays) involves exposure to radiation.
- Participants taking an anti-platelet agent (e.g. aspirin) may, at the discretion of the site clinician, be discontinued from that agent for a period of time prior to lumbar puncture and/or continue off agent for a period of time post LP. Participants who are taking anticoagulants (e.g. warfarin (Coumadin) and/or dabigatran (Pradaxa)) may not undergo an LP and are not suitable to participate in this study.
- Each study participant or a person designated to speak for them will be contacted by phone one day after the LP to confirm participant well-being and to query about any adverse events.
- Identify a physician (e.g., anesthesiologist) able to perform a blood patch for any participant who experiences a post lumbar puncture headache. Find out ahead of time who to call to schedule and perform a blood patch at your center, should the need arise. Ensure billing procedures are in place ahead of time.
- Ensure you have at least two “Lumbar Puncture Tray Kits” and sufficient “CSF Supplemental Supply Kit” provisions on hand prior to scheduling an LP visit. Also ensure adequate site-provided supplies (see above), including pelleted dry ice, are available. Check expiration dates on all supplies, especially lidocaine.

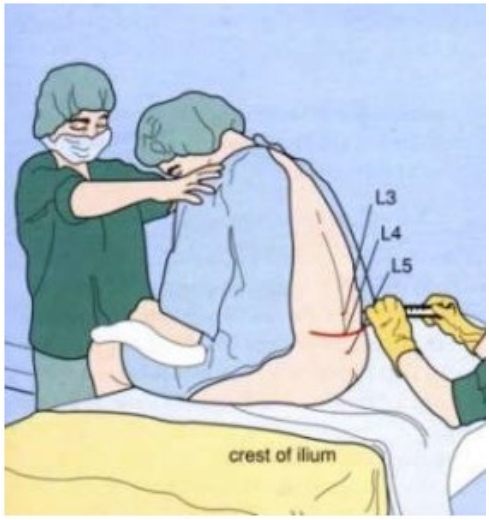
8.1 Scheduling the LP

All LPs should be performed in the morning if possible. Availability of staff and facilities for next day blood patch should be considered when scheduling LPs. CSF amyloid levels can vary depending upon the time of day the sample is collected. It is important for the time of day of collection to remain consistent across study visits.

The LP should be rescheduled if the participant does not feel well or is febrile.

8.2 Performing the LP

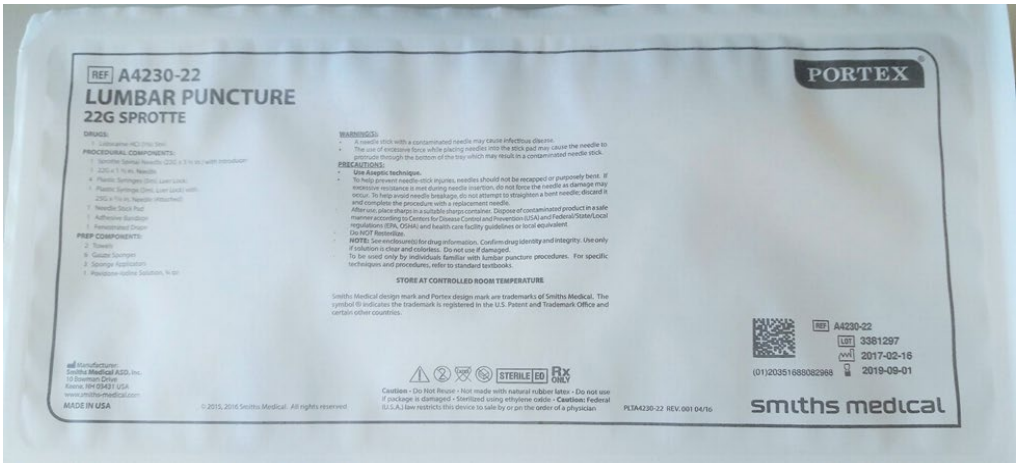
The recommended position is sitting with curved back and head down. For comfort, a stool may be used to prop up the feet and legs. The same position should be used at follow-up LPs. It is critical to try to optimize positioning, and usually requires an assistant. Other positions and needles are allowed (e.g., when using fluoroscopy) but this should be recorded in the ADCFB Specimen Collection And Processing Form. A pillow may be placed under the head for comfort.



On the bedside table nearest where the person performing the lumbar puncture will sit, place a pair of sterile gloves (in their packaging) and a blue pad. Remove the contents of the lumbar puncture tray from the outer plastic packaging, leaving the contents wrapped in their sterile drape. Leave everything wrapped until the person performing the lumbar puncture is seated.

Feel the outside of the lumbar puncture kit (still wrapped up) to determine which end contains the spongy swabs. Turn this end toward the person performing the lumbar puncture and begin un-wrapping the kit.

Lumbar Puncture Tray Kit Images



Exterior of LP Tray provided by NCRAD containing the 22 gauge Sprotte Needle with Introducer.

Close up of Sprotte Spinal Needle (22 gauge x 3 ½ in.) with Introducer

(24 gauge is equivalent but with lavender top needle)

Interior of LP Tray Provided by NCRAD

TOUCH ONLY THE OUTSIDE OF THE PAPER WRAPPER. When you grab an edge to unfold it, touch only the folded under portions of the outside of the wrapper. Also, don't let the outside of the wrapper touch any part of the inside.

- If you touch any part of the paper wrapper, or if any non-sterile object outside of the wrapper touches any part of the inside of the wrapper, throw the kit away and start over.
- If you are in any doubt as to whether the inside of the wrapper has been touched, throw the kit away and start over.

Cleaning the Lumbar Puncture Site
The lumbar puncture site is cleaned with Povidone-Iodine Topical Solution according to best standard medical practices.

Once the kit is successfully unwrapped, open the bottle of Povidone-Iodine Topical Solution somewhere away from the kit. Use an alcohol swab to remove any loose chunks of dried material off of the bottle top. You don't want anything to fall onto the open and sterile lumbar puncture kit. Pour enough Povidone-Iodine Topical Solution into the prep well to cover the bottom, about ¼ inch deep.

Maintaining the Sterile Field
An important aspect of assisting with a successful lumbar puncture is keeping the field sterile. If there are a number of staff members in the room, please be sure they do not accidentally contaminate the sterile field. Once the person performing the lumbar puncture has donned sterile gloves, additional help may be needed to obtain or un-wrap any new tubes, needles, or supplies.

Unwrapping the Sterile 15 and 50 ml Conical Tubes
Note that the 15 ml and 50 ml tubes into which CSF is collected and transferred come individually wrapped and are sterile inside and out. These wrappers should be peeled open by an assistant (not touching the tube) and the tube carefully dropped onto the LP tray or elsewhere in the sterile field in a manner that avoids contamination. Any additional needles or other individually-wrapped sterile items can be handled the same way.

- Do not drop any packaging onto the tray or sterile field.
- Do not let the item touch the outside of the packaging on its way to the tray.

Lidocaine, Syringe with Needle, Gauze Pads
Anesthesia is usually achieved within 2 minutes after injecting the lidocaine. Occasionally, the person performing the lumbar puncture will need to use more lidocaine to numb up a particular spot, or they may need to move to another spot entirely.

Hold the lidocaine bottle upside down and at a slight angle toward the person performing the lumbar puncture so that they can plunge the needle into the bottle and extract some lidocaine without touching you or the bottle. Use two hands to stabilize the bottle. If the person performing the LP requires additional sterile gauze, open the gauze pad the same way as the syringe and needle, by holding open the package so the person performing the lumbar puncture can grab the gauze without touching you or the package.

General CSF Collection Methods
LPs for CSF collection should be performed using a small caliber atraumatic needle. CSF should be obtained via gravity flow using the 22 gauge Sprotte needle, although aspiration through this or smaller needles is allowable. Prior approval from the Clinical Core is required before the aspiration method can be utilized. Sites must designate the method of CSF collection for data tracking purposes. It is recommended that CSF be obtained from participants in a sitting position. Alternate needles, positions or methods (e.g., use of fluoroscopy) should be noted in Comments section of the ADCFB Specimen Collection And Processing Form.

Collection of CSF by Gravity
After the spinal needle enters the L3-4 or adjacent intrathecal space and the stylet is withdrawn, CSF should flow freely. **Discard first 1-2 ml of CSF if blood tinged. If not blood tinged, collect first 1-2 ml of CSF into a 15 ml conical tube and pipette into the yellow cap cryovial for local lab. Collect 15-20 ml CSF total into the remaining two 15 ml conical tubes.**

Reminder: If the CSF is blood-tinged, the first 1-2 ml of CSF should be discarded (or more if needed) to clear the blood before collecting the 15-20 ml of CSF for research use. If 15 ml is not obtained and provided to NCRAD, document the reason for under-collection in the comments section of the ADCFB Specimen Collection And Processing Form.

Up to 20 ml of CSF can be collected for the ADCFB protocol. Any additional CSF collected will require a separate informed consent document that is connected to a specific protocol. NCRAD recommends to avoid exceeding a total collection volume of 40 ml.

Washcloths, Band-Aids, and Clean Up
After the person performing the lumbar puncture collects the last of the CSF, remove the needle and introducer and wash the Povidone-Iodine Topical Solution off the participant. A warm, wet washcloth can be used. A Band- Aid should be applied to the puncture site. The participant should lie flat for 30-60 minutes. Next, discard the LP kit following local guidelines, and dispose of sharp components in an appropriate sharps container.

Suggested management of post-lumbar puncture headache
Classic post-lumbar puncture (low pressure) headache typically begins 24-48 hours after dural puncture, and the headache is worse when the participant is upright (sits or stands) and improves when the participant is recumbent with the head **no higher** than the spinal cord.

Safety and comfort of the LP is maximized by the use of atraumatic needles. The protocol requires use of a 22 gauge Sprotte needle. Lumbar puncture is a standard procedure for collection of CSF but may be associated with pain during the performance of the procedure, comparable to the level of pain experienced during a blood draw. This is usually temporary and confined to the lower back. A persistent low-pressure headache may develop after lumbar puncture, probably due to leakage of CSF. If a post-LP headache persists it may need additional treatment, e.g. with fluids and analgesics. Uncommonly, a blood patch (injection of some of the participant's blood to patch the CSF leak) may be needed.

Prevention: Use of a small gauge and atraumatic needle with careful technique are helpful in preventing post-lumbar puncture headache. Having the participant refrain from exercise or strenuous activities (especially heavy lifting) and staying well-hydrated for 24 hours after the LP may minimize the chance of a lumbar puncture headache.

Treatment of headache after a lumbar puncture:

- Limit physical activity as much as possible for at least 24 hours post-procedure.
- Increase oral fluid intake. Caffeine may be helpful.
- Routine analgesics such as acetaminophen may be used.

Post-lumbar puncture headache often resolves with the above treatment. If the headache persists after 24 hours of this management, it will likely require a blood patch. A blood patch *typically* relieves the headache instantly.

8.3 Step by Step Summary of CSF Collection Procedure

1. Ensure all samples collected are appropriately labeled.

2. Open ADCFB Specimen Collection And Processing Form.

3. Confirm all supplies are available.

4. Label the thirteen orange-capped cryovials and one blue-capped cryovial with provided CSF Aliquot Labels. Do **NOT** open and label the 15 ml and 50 ml tubes that will be kept sterile to collect the CSF.

5. Pre-cool the centrifuge and pre-cool all fourteen labeled cryovials on wet ice. Do **NOT** pre-cool the 15 ml and 50 ml tubes that will be kept sterile to collect the CSF.

6. Measure vitals (participant lying down).

7. Record the time of LP and associated information in the ADCFB Specimen Collection And Processing Form.

8. Collect 15-20 ml CSF at the L3/L4 position (or adjacent position) using a 22 gauge Sprotte spinal needle via gravity flow with participant in upright position (or document alternate method in the ADCFB Specimen Collection And Processing Form) following these steps:
    a. Collect initial 1-2 ml (if bloody, collect CSF until cleared of blood) using the 15 ml conical tube. If not bloody, transfer first 1-2 ml into yellow-capped cryovial for local lab.
    b. Collect an additional 15-20 ml CSF into the unlabeled and sterile 15 ml polypropylene tubes from the "CSF Supply Kit". 15 ml is the required minimum.
    c. If using aspiration, use **ONLY** the polypropylene syringes included in the "Lumbar Puncture Collection Kit" and transfer directly into the unlabeled and sterile 15 ml polypropylene tube from the "CSF Supply Kit". There are four 6 ml Luer lock polypropylene syringes in the "Lumbar Puncture Collection Kit." Note if aspiration method was used in the ADCFB Specimen Collection And Processing Form.

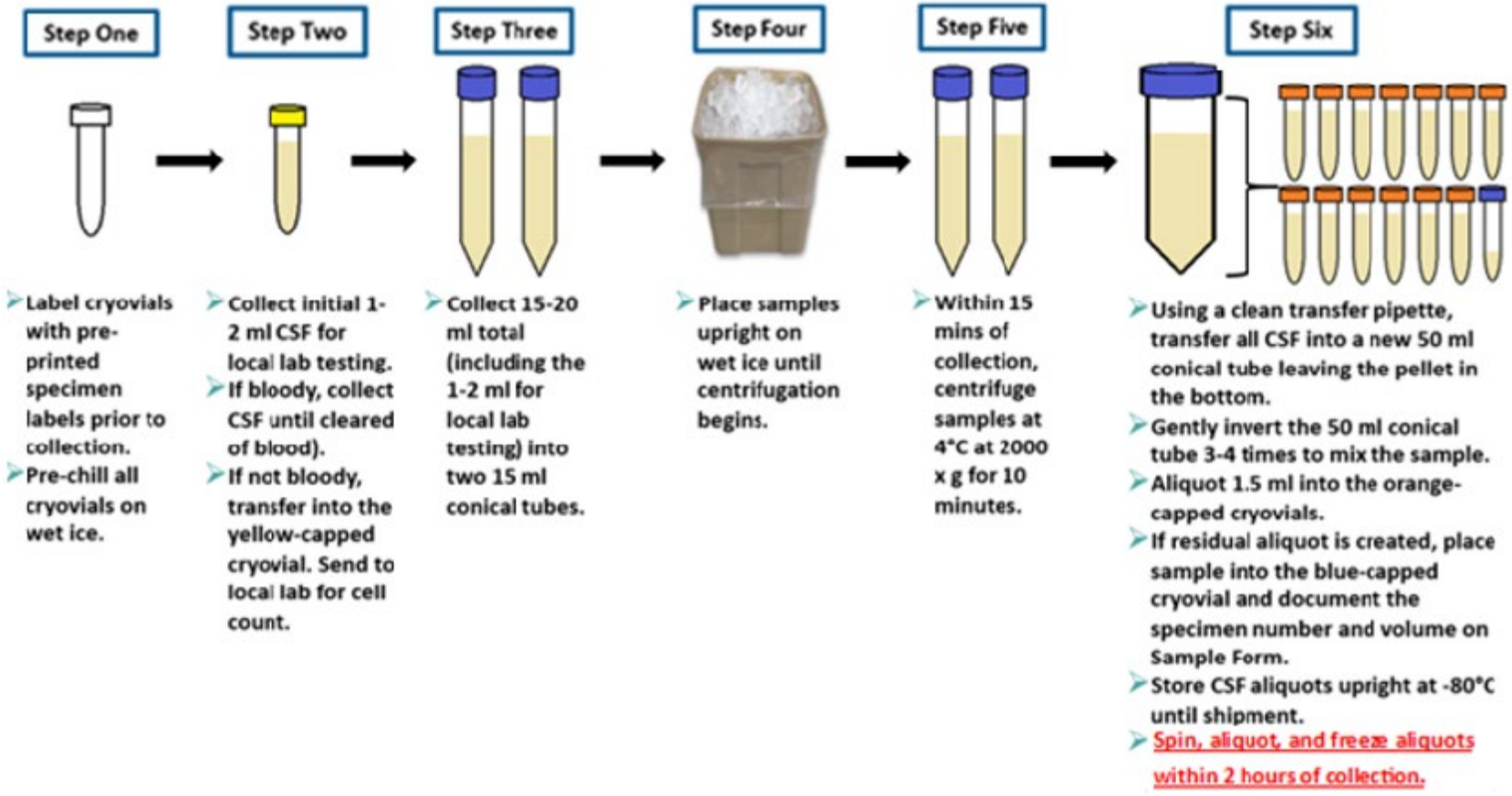
9. As one person takes the immediate post procedure vital signs, a second person should process the CSF as follows:
    a. Place samples upright on wet ice and ensure samples are kept on wet ice for the entire time prior to processing. Preferably within 15 minutes of collection, centrifuge briefly at low speed (2000 x g, 10 min, 4°C) to pellet any cellular debris.
    b. Using a clean transfer pipette, transfer CSF from both 15 ml conical tubes into a 50 ml conical tube, leaving the debris at the bottom of each 15 ml centrifuged tube. Gently invert the 50 ml conical tube 3-4 times to mix the sample.
    c. Aliquot 1.5 ml volumes into the orange-capped cryovials. If a residual aliquot is created, aliquot into blue-capped cryovial. Document specimen number and volume in the ADCFB Specimen Collection And Processing Form.
    d. Within 2 hours of CSF collection, samples need to be spun, aliquoted and in the freezer. Store CSF aliquots at -80°C until shipment. Record time of freezing in the ADCFB Specimen Collection And Processing Form.

10. Provide food and drink to participant (participant may lay flat to minimize the chance of a post-LP headache).

11. Place the labeled cryovials in the 25 cell cryobox and place on pelleted dry ice. Transfer to -80°C Freezer when possible. Store all samples at -80°C until shipped to NCRAD on pelleted dry ice. Record time aliquots placed in freezer and storage temperature of freezer in the ADCFB Specimen Collection And Processing Form.

CSF Aliquots (up to 14 possible)

CSF Preparation (15-20 ml total)



Check expiration dates of supplies before collection to make sure they are not expired!

9.0 Packaging & Shipping Instructions

ALL study personnel responsible for shipping should be certified in biospecimen shipping. If you have difficulty finding biospecimen shipping training, please notify a NCRAD coordinator.

In addition to tracking and reconciliation of samples, the condition and number 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 frozen samples are packed with sufficient amounts of pelleted dry ice to avoid thawing in the shipment process.

9.1 Shipping Notifications

Complete the NCRAD Specimen Shipment Notification Form for each package going to NCRAD on the day of shipment. The notification survey collects the following information:

1. Study: Select ADCFB from the drop down.
2. Name of study contact completing form, include first and last name.
3. Email of study contact completing form.
4. Emails of additional contacts to be notified about the shipment's status (up to 3).
5. Courier: Select UPS for ADCFB.
6. Shipment Tracking Number
7. Shipment type: Frozen or ambient
8. Sample type(s) included in the shipment.
9. Kit Numbers included in the package (up to 8). If less than 8 kits are in the package, enter "N/A" in the remaining spaces.
10. Participant IDs associated with the kits included in the shipment (up to 8). If less than 8 patients' samples are in the package, enter "N/A" in the remaining spaces.
11. PDF file of sample forms included in the shipment. If there are multiple forms they can be combined into one PDF or into a zipped file prior to upload.
12. Date of the shipment.

Completion of this survey automatically notifies the NCRAD email address (alzstudy@iu.edu) and the selected study's Coordinator.

9.2 Ambient Packaging Instructions

*****Important Note*****
AMBIENT SAMPLES MUST BE SHIPPED MONDAY-THURSDAY ONLY!

Ambient PBMC samples must be shipped the day of blood draw, so do not draw on Fridays.

Ambient sodium heparin (green-top) sample shipments should be considered as Category B UN3373 and as such must be tripled packaged and compliant with the IATA Packing Instructions 650. *See the Latest Edition of the IATA Regulations for complete documentation.*

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.

*** Ambient Shipping Packing and Labeling Guidelines ***

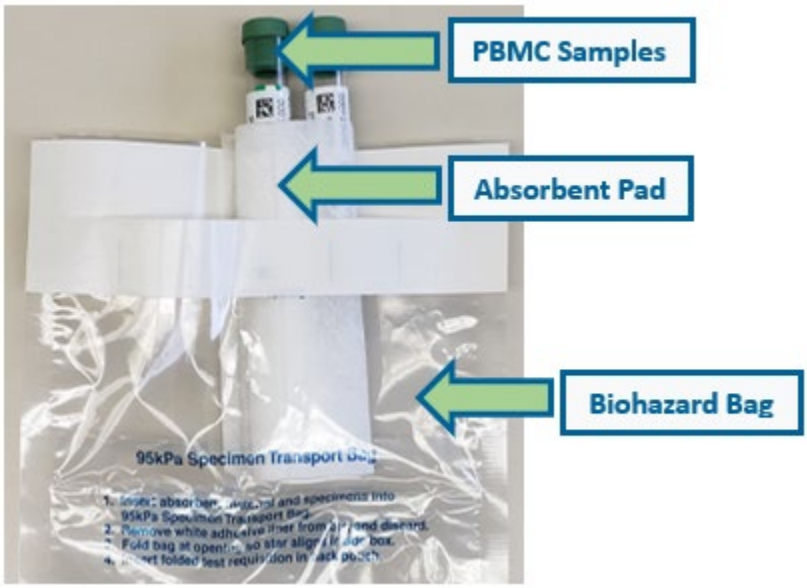
- The primary receptacle (sodium heparin tube) must be leak proof and must not contain more than 10 ml total.
- The secondary packaging (small biohazard bag) must be leak proof.
- Absorbent material must be placed between the primary receptacle and the secondary packaging (small biohazard bag). The absorbent 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

9.2.1 NCRAD Packaging Instructions—Ambient Shipments

1. Place refrigerant pack in the freezer 24 hours prior to shipment.

2. Notify NCRAD of shipment by completing the NCRAD Specimen Shipment Notification Form.

3. Place filled and labeled sodium heparin (green-top) tubes into the plastic biohazard bag with absorbent sheet. If absorbent tube sleeve is included, place tubes in sleeve slots then place sleeve with tubes in the biohazard bag.



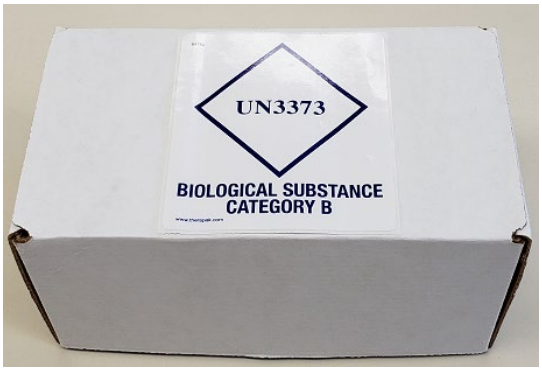
4. Remove as much air as possible from the plastic biohazard bag and seal the bag according to the directions printed on the bag.

5. Place the refrigerant pack into the cooler on top of the filled biohazard bag.

6. Place the lid onto the cooler.

7. Place a hardcopy of the paperwork for each kit included in the package on top of the cooler lid along with a completed list of contents card. Acceptable paperwork includes:
   a. ADCFB Collection and Processing Form – Condensed
   b. Appendix C: Blood Sample and Shipment Notification Form

8. Close the shipping box. Label the outside of the cardboard box with the enclosed UN3373 (Biological Substance Category B) label.



9. Place the closed, labeled shipping box within a UPS Laboratory Pak. **Seal the UPS Laboratory Pak.**

10. Place UPS return airbill on the sealed UPS Laboratory Pak.

11. Use UPS tracking to ensure the delivery occurs as scheduled and is received by NCRAD.

9.3 Frozen Packaging Instructions

The most important issue for shipping is to maintain the temperature of the samples. The frozen samples must never thaw; not even the outside of the tubes should be allowed to defrost. This is best accomplished by making sure the Styrofoam container is filled completely with pelleted dry ice.

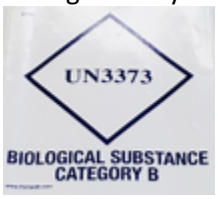
*****Important Note*****
FROZEN SAMPLES MUST BE SHIPPED MONDAY-WEDNESDAY ONLY!

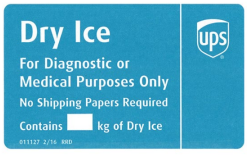
Specimens being shipped to NCRAD should be considered as Category B UN3373 specimens and as such must be tripled packaged and compliant with IATA Packing Instructions 650. *See the Latest Edition of the IATA Regulations for complete documentation.*

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.

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

- The primary receptacle (cryovial) 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 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
  - ✓ UPS Dry Ice label and net weight of dry ice contained.





9.3.1 NCRAD Packaging Instructions – Frozen Shipments

1. Notify NCRAD of shipment by completing the NCRAD Specimen Shipment Notification Form.
    a. Attach sample paperwork for each kit included in the package to the Notification Form, acceptable paperwork includes:
        i. ADCFB Collection and Processing Form – Condensed
        ii. Appendix C: Blood Sample and Shipment Notification Form
        iii. Appendix D: CSF Sample and Shipment Notification Form

2. Place the cryovial boxes containing frozen samples into a biohazard bag.

3. As the cryovial box is placed in the plastic biohazard bag, do NOT remove the absorbent material found in the bag. Seal according to the instructions on the bag.

4. Place approximately 2-3 inches of pelleted dry ice in the bottom of the Styrofoam shipping container.

5. Place the biohazard bags into the provided Styrofoam-lined shipping container on top of the pelleted dry ice. Please ensure that cryovial boxes are placed so the cryovials are upright in the shipping container.
    a. Do NOT overpack frozen shippers. Small shippers have capacity for a **MAXIMUM of 3 kits**. Large shippers have capacity for a **MAXIMUM of 8 kits.**
    b. NCRAD's frozen shipping kits are designed to maximize temperature stable transit time. NCRAD is not responsible for sample loss resulting from overpacked or under iced shippers.

6. Fully cover the biohazard bags containing the cryovial boxes tubes with approximately 2 inches of pelleted dry ice.

7. After the samples have been placed into the shipping container, completely fill the inner Styrofoam with dry ice pellets to ensure the frozen state of the specimens during transit.

8. Replace the lid on the Styrofoam carton.

9. Include a hardcopy of the paperwork for each kit included in the package on top of the Styrofoam lid (do not place paperwork in biohazard bags or over dry ice), acceptable paperwork includes:
    c. ADCFB Collection and Processing Form – Condensed
    d. Appendix C: Blood Sample and Shipment Notification Form
    e. Appendix D: CSF Sample and Shipment Notification Form

10. Close and seal the outer cardboard shipping carton with packing tape.

11. Complete the UPS Dry Ice Label with the following information:
    b. Net weight of dry ice in kg (must match amount on the airbill)
    c. Do not cover any part of this label with other stickers, including preprinted address labels.

12. Apply all provided warning labels and UPS return airbill to the outside of package, taking care not to overlap labels.

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

Complete the required fields on the UPS Dry Ice label or UPS may reject or return your package.

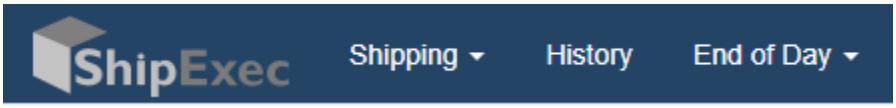
13. If possible, hold packaged samples in -80°C freezer until time of UPS pick-up/drop-off. If storage in a -80°C freezer until UPS pick-up is not possible, package samples no more than 4 hours before the expected pick-up time.

14. Use UPS tracking to ensure the delivery occurs as scheduled and is received by NCRAD. Include tracking information when completing the NCRAD Specimen Shipment Notification Form.

9.4 Ambient and Frozen Shipping Instructions

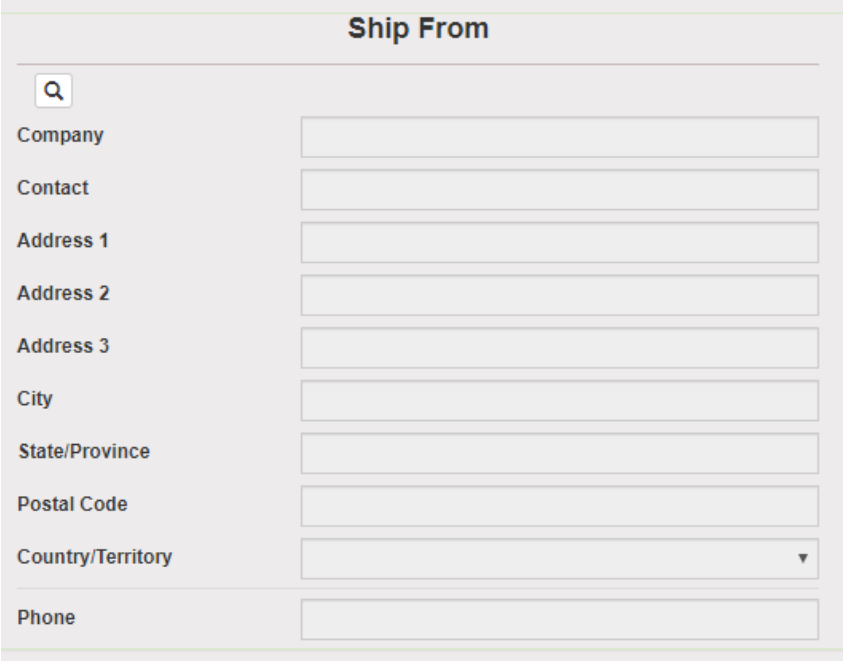
1. Log into the ShipExec Thin Client at kits.iu.edu/UPS.
    a. If a new user or contact needs access, please reach out to your study contact for access.

2. Click “Shipping” at the top of the page and select “Shipping and Rating”.



3. Select your study from the “Study Group” drop down on the right side of the main screen. Choosing your study will automatically filter the address book to only addresses within this study.

4. Click on the magnifying glass icon in the “Ship From” section to search for your shipping address.



    a. Search by Company (site), Contact (name), or Address 1 (first line of your site’s street address). Click Search.
    b. Click Select to the left of the correct contact information.

5. Verify that both the shipping information AND study reference are correct for this shipment.
    a. If wrong study contact or study reference, click Reset in the bottom right of the screen to research for the correct information.

6. Enter Package Information
    a. **Ambient shipments**
        i. Enter the total weight of your package in the “Weight” field and leave the “Dry Ice Weight” field empty.
    b. **Frozen shipments**
        i. Enter the total weight of your package in the “Weight” field.
        ii. Enter the dry ice weight in the “Dry Ice Weight” field.
        iii. If the “Dry Ice Weight” field is higher than the “Weight” field, you will receive an error message after clicking Ship and need to reenter these values.
    c. Click Ship in the bottom right of the page when complete.

7. If your site does not already have a daily UPS pickup, you can schedule one here.
    a. Click the blue Pickup Request button. Enter the earliest pickup time and latest pickup time in 24-hr format.
    b. Give a name & phone number of someone who the UPS driver can call if having issues finding the package.
    c. Give the Floor and Room Number (if needed) to be as descriptive as possible where this package needs to be picked up from. Click Save.

8. Print the airbill that is automatically downloaded.
    a. To reprint airbill, click History at the top left of the page.
        i. Shipments created from the user that day will automatically populate. If shipments from a previous day need to be located, search by ship date.
        ii. Locate the correct shipment, and click on the printer icon to the left of the tracking number under “Action” to reprint the airbill
        iii. Click print icon on right side of the tracking number line.

9. Fold airbill, and place inside plastic UPS sleeve.

10. Peel the back off of the UPS sleeve and stick the sleeve to the package top. Ensure that sleeve does not cover any warning labels (e.g. dry ice label) or overlap taped seams.

10.0 ADCFB Specimen Collection And Processing Form

Completion of the ADCFB Specimen Collection And Processing Form is required in order to expedite data entry for purposes of running plasma biomarkers. This survey replaces both Appendix C: Blood Sample and Shipment Notification Form and Appendix D: CSF Sample and Shipment Notification Form. Paper forms can be completed during processing as needed but the online ADCFB Specimen Collection And Processing Form must be completed prior to sample shipment. Failure to complete the online processing form may significantly delay receipt of biomarker data.

The ADCFB Specimen Collection And Processing Form consists of three main parts:

1. Site and Patient Information
    a. Study Site
    b. Site contact information
    c. ADRC Patient ID
    d. GUID (if collected)
    e. NACC ID (if known at time of collection)
    f. Sex
    g. Year of Birth
2. Visit Information
    a. NACC Visit
    b. Kit Number
    c. Visit Type (Blood or CSF)
3. Collection Information
    a. All blood or CSF Processing information collected in the historical Blood Sample and Shipment Notification and CSF Sample and Shipment Notification forms.

At the end of the ADCFB Specimen Collection And Processing Form click “Continue to Review” to review a condensed form and check the entered information. If everything looks correct, click “Submit” to send the completed form to NCRAD. Once the survey has been submitted to NCRAD it can no longer be edited by external parties. Email the NCRAD ADCFB Coordinator (ssteidel@iu.edu) to request any necessary corrections.

Alternatively, click “Save & Return Later” to save the survey and receive a link to access and complete the survey later. These surveys are considered incomplete. The Kit Number and Participant ID associated with the survey is required to access the survey again later. This method can be used to allow multiple staff members to complete the form if needed.

Once submitted the site contact included on the form will receive two emails:

1. ADCFB Collection and Processing Form Received (Kit 1234567)

a. This email confirms receipt of the form and includes a copy of the full survey.

2. ADCFB Collection and Processing Form - Condensed (Kit 1234567)
   a. This email includes a copy of a condensed one-page version of the survey which can be included in the NCRAD Specimen Shipment Notification Form and printed to include with the sample shipment.

11.0 Data Reconciliation

The ADCFB Specimen Collection And Processing Form or an equivalent Sample and Shipment Notification Form must be completed on the day that samples are collected because they include information that will be used to reconcile sample collection and receipt, as well as information essential to future analyses.

NCRAD will collaborate with the data team at NACC to reconcile information captured in the NACC database compared to samples received and logged at NCRAD. Additional discrepancies may be sent directly to the center staff to reconcile.

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

- Incorrect samples collected and shipped.
- Damaged or incorrectly prepared samples.
- Unlabeled samples, samples labeled with incomplete information, or mislabeled samples.
- Discrepant information documented in the ADCFB Specimen Collection And Processing Form or on the Blood or CSF Sample and Shipment Notification Forms and logged at NCRAD compared to information entered into the NACC database.

12.0 Appendices

Appendix A: GUID Demographics Form

Please be certain to collect the following demographic information to generate a Global Unique Identifier. **Do NOT** return this information to NCRAD. Only send the GUID to NCRAD.

1. Compete legal given (first) name of participant at birth:_____
2. Complete additional (middle) name or names at birth: _____
3. Complete legal family (last) name of participant at birth:_____
4. Suffix: _____
5. Date of Birth: _____
6. Name of city/municipality in which participant was born: _____
7. Country of birth: _____

Appendix B: Rate of Centrifuge Worksheet

Please complete and return this form by 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:

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

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 at alzstudy@iu.edu

Appendix C: Blood Sample and Shipment Notification Form

Please complete the NCRAD Specimen Shipment Notification Form before shipment.

To: Kelley Faber Email: alzstudy@iu.edu Phone: 1-800-526-2839

From:_____ **UPS tracking #:**1Z976R8W84

Phone:_____ **Email:**_____

Study: ADCFB **Sex:** ☐M ☐F **Year of Birth:** _____
Site ID:_____ **PT ID:**_____
GUID:_____
NACC Visit: _____

KIT BARCODE

Blood Collection:

Date of Draw: _____ [MMDDYY]	Time of Draw: _____ [HHMM]
Date participant last ate: _____[MMDDYY]	Time participant last ate: _____ [HHMM]

PBMC (NaHep Tubes) ☐N/A

#1	Specimen Number **(Last four digits)**:	_____	Original volume drawn:	_____ml
#2	Specimen Number **(Last four digits)**:	_____	Original volume drawn:	_____ml

Blood Processing: **Plasma & Buffy Coat (EDTA Tube)**

EDTA #1 specimen number **(Last four digits):**	_____	Original blood volume of EDTA #1:	_____ mL
EDTA #2 specimen number **(Last four digits):**	_____ ☐N/A	Original blood volume of EDTA #2:	_____ mL ☐N/A
EDTA #3 specimen number **(Last four digits):**	_____ ☐N/A	Original blood volume of EDTA #3:	_____ mL ☐N/A
Time spin started:	_____ [HHMM]	Duration of centrifuge:	_____ mins
Temp of centrifuge:	_____ °C	Rate of centrifuge:	_____ x g
Time aliquoted:	_____ [HHMM]	Number of 1.5 mL plasma aliquots created (purple cap):	_____
Volume of BAL plasma aliquot (0.75 mL in yellow cap):	_____ mL	Specimen number of BAL plasma aliquot **(Last four digits)**:	_____
Volume of residual plasma aliquot (less than 1.5 mL in blue cap):	_____ mL ☐N/A	Specimen number of residual plasma aliquot **(Last four digits)**:	_____ ☐N/A
Buffy coat #1 specimen number **(Last four digits):**	_____	Buffy coat #1 volume:	_____ mL
Buffy coat #2 specimen number **(Last four digits):**	_____ ☐N/A	Buffy coat #2 volume:	_____ mL ☐N/A
Buffy coat #3 specimen number **(Last four digits):**	_____ ☐N/A	Buffy coat #3 volume:	_____ mL ☐N/A
Time aliquots frozen:	_____ [HHMM]	Storage temperature of freezer:	_____ °C

Notes:_____

Appendix D: CSF Sample and Shipment Notification Form

Please complete the NCRAD Specimen Shipment Notification Form before shipment.

To: Kelley Faber Email: alzstudy@iu.edu Phone: 1-800-526-2839

From:_____ **UPS tracking #:**1Z976R8W84

Phone:_____ **Email:**_____

Study: ADCFB **Sex:** ☐M ☐F **Year of Birth:** _____

Site ID: _____ **PT ID**: _____

GUID: _____

NACC Visit: _____

KIT BARCODE

CSF Collection:

Date of Draw: _____ [MMDDYY]	Time of Draw: _____ [HHMM]
Date participant last ate: _____[MMDDYY]	Time participant last ate: _____ [HHMM]
Collection process: ☐ Gravitational **OR** ☐ Pull	Needle used to collect CSF: ☐ 20g Quincke ☐ 22g Sprotte ☐ 22g Quincke ☐ 24g Sprotte ☐ 25g Quincke ☐ Other *(please specify)*: _____

CSF Processing:

Time spin started:	_____ [HHMM]
Duration of centrifuge:	_____ mins
Temp of centrifuge:	_____ °C
Rate of centrifuge:	_____ x g
Total amount of CSF collected (mL):	_____ mL
Time aliquoted:	_____ [HHMM]
# of 1.5 mL CSF aliquots created: **(Orange-capped cryovial)**	_____
If applicable, volume of CSF residual aliquot (less than 1.5 mL): **(Blue-capped cryovial)**	_____ mL
If applicable, specimen number of residual aliquot: **(Last four digits)**	_____
Time aliquots frozen:	_____ [HHMM]
Storage temperature of freezer:	_____ °C

Notes: _____