

## CAPTURE ALS



# BIOSAMPLE COLLECTION, PROCESSING, STORAGE, SHIPMENT

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SOP01EN03


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## 1. SCOPE AND APPLICABILITY

This SOP describes the processes for the labelling, collection, processing, shipment, and storage of CAPTURE ALS biosamples including blood and cerebrospinal fluid (CSF).

## 2. SUMMARY OF METHOD

Following participant consent, blood, and optional CSF will be collected by qualified medical professionals according to the schedule outlined in the CAPTURE ALS protocol and following local site SOPs.

Blood: Blood will be processed at the site of collection to isolate serum and plasma, which will be aliquoted, and batch shipped to the C-BIG Repository for long-term storage. In general, blood will be shipped to and processed by the C-BIG Repository to isolate DNA, and PBMCs and for long-term storage of RNA.

CSF: 1-2mL of CSF will be sent to the site's local laboratory for analysis of cell count, protein, and glucose. The remaining CSF will be processed and aliquoted at the site of collection and batch shipped to the C-BIG Repository for long-term storage.

## 3. DEFINITIONS

C-BIG	Clinical Biological Imaging and Genetic Repository
CSF	Cerebrospinal Fluid
DNA	Deoxyribonucleic acid
DOT	Department of Transportation
EDTA	Ethylenediaminetetraacetic acid
g	Gravity
GTT	Green Top Blood Collection Tube (Sodium Heparin)
IATA	International Air Transport Association
LIMS	Laboratory Information Management System
LORIS	Longitudinal Online Research and Imaging System
MD	Medical Doctor
NCCLS	National Committee for Clinical Laboratory Standards
OSHA	Occupational Safety and Health Administration
PBMCs	Peripheral blood mononuclear cells
PTT	Purple Top Blood Collection Tube
RBC	Red blood cells
RNA	Ribonucleic acid
RTT	Red Top Blood Collection Tube

## 4. PERSONNEL QUALIFICATIONS/RESPONSIBILITIES

This SOP concerns all personnel who will be delegated to process human biosamples for CAPTURE ALS. All personnel processing specimens must be currently certified in the transportation of dangerous goods.

## 5. CAUTIONS

- All biosamples derived from a human source are considered to be biohazardous. Use appropriate precautions when working with such samples (i.e. personal protection equipment such as gloves, lab coat and safety glasses). All waste (samples and related contact materials) must be placed in marked biohazardous waste containers and disposed of under hospital or institutional guidelines.
- Ensure that informed consent is obtained prior to participant biosample collection.
- Follow your facility's SOPs for biosample collection.
- Blood collection at each visit must not exceed 12 tubes of blood which is equivalent to 8 tablespoons or 120mL.
- CSF collection at each visit must not exceed 16mL.
- Best results are obtained when samples are processed promptly.

## 6. EQUIPMENT AND SUPPLIES

The equipment and supplies listed below are recommendations only and may be substituted by alternative/equivalent product more suitable for site-specific tasks or procedures.

### 6.1. Equipment

- Room temperature centrifuge with swinging bucket (2000-2500g)
- Refrigerated centrifuge with swinging bucket (250g)
- -20°C freezer
- -80°C freezer
- Micropipettes (100-1000µL) and tips
  - **Note:** if a pipette is not available, sterile transfer pipettes may be used
- Laser printer
- Windows computer with Microsoft Office and working internet
- Barcode scanner
  - Suggested: Eyoyo Wired USB 2D Barcode Scanner, Model EY-H2  
[https://www.amazon.ca/Eyoyo-Handheld-Convenience-Supermarket-Warehouse/dp/B088QV215Y/ref=pd\\_lpo\\_1?pd\\_rd\\_i=B088QV215Y&psc=1](https://www.amazon.ca/Eyoyo-Handheld-Convenience-Supermarket-Warehouse/dp/B088QV215Y/ref=pd_lpo_1?pd_rd_i=B088QV215Y&psc=1)

### 6.2. Supplies

CAPTURE ALS *suggests* the following supplies for the local collection, processing, storage, and shipment of all CAPTURE ALS biosamples.

Supply Name	Supply Description	Suggested Vendor	Item/ Catalog #	Comments
Blood Collection Materials	N/A	At site's discretion	N/A	alcohol wipes, needles, gauze, bandaids etc

Cryovials, 1mL, conical bottom	Nunc Biobanking and cell culture cryogenic tube 1mL; polypropylene; screw cap; self-standing	ThermoFisher	366656	10 cryovials = plasma 15 cryovials = serum 45 cryovials = CSF
Sterile Transfer Pipettes	Corning™ Falcon™ Plastic Disposable Transfer Pipet; Individually Wrapped; Sterile 3mL; 1,2mL graduations	Fisher Scientific	13-669-12	To aliquot samples
15mL Tube	15mL polypropylene tube; graduation; screw cap	Sarstedt	62.554.205	1 for processing of each, serum, plasma, CSF
Cryogenic labels	Fisherbrand Micryo Label; Laser Printer Sheets; 1.2 x 2.5 cm, -196°C to 121°C	Fisher Scientific	15-930-A	1 label per vacutainer and cryovial Use FISHC6 label template
<b>SERUM COLLECTION</b>				
BD Vacutainer™ venous blood collection tube, red top (RTT)	10.0mL; Glass; Conventional Closure; Red Cap; Silicone Coated	Fisher Scientific	02-685-A (BD 366430)	2 tubes per serum sample
<b>PMBC COLLECTION</b>				
BD Vacutainer™ blood collection tube with Sodium Heparin, green top (GTT)	10mL; Glass; Conventional Closure; Green cap; Sodium Heparin	Fisher Scientific	02-685-3B (BD 366480)	5 tubes per PBMC sample
<b>PLASMA AND DNA COLLECTION</b>				
BD Vacutainer™ blood collection tube with K <sub>2</sub> EDTA, purple top (PTT)	10mL; Plastic; Hemogard Closure; Lavender Cap; K <sub>2</sub> EDTA	Fisher Scientific	02-657-32 (BD 366643)	2 tubes for DNA 1 tube for plasma sample
<b>RNA COLLECTION</b>				
PAXgene blood RNA tube	2.5mL; Plastic; Hemogard Closure; Red Cap; RNA stabilization additive	BD	BD 762165	2 tubes per RNA sample
<b>CSF COLLECTION</b>				
Lumbar Puncture Tray, preferred needle, additional supplies	N/A	At site's discretion		1 kit per LP
15mL Tube	15mL polypropylene tube; graduation; screw cap	Sarstedt	62.554.205	2 per CSF collection
<b>SHIPPING MATERIALS</b>				
Parafilm, Elastics, Packing Tape	N/A	At site's discretion	N/A	
Thermo Scientific Cryoboxes	Cryoboxes, Chipboard, White, holds 81 vials, 132 x 132 x 51mm	Fisher Scientific	12-565-182	Holds 81 cryovials split participant samples across 2 boxes
Absorbent Materials	Absorbent Sheet; 3 x 6 in.; Absorbs up to 90mL	Fisher Scientific	22-130-040	Utilize per IATA standards

Dry Ice Label	Labels, Dry ice; Therapak; 5.5 x 5.5 in.; Complies with DOT (49 CFR 173.217) and IATA	Fisher Scientific	22-130-065	1 per shipping box
UN 3373 Biological Substance Category B Labels	8 x 4 in.; for shipping biological substances by air; complies with IATA regulations	Fisher Scientific	22-130-069	1 per shipping box
Biohazard Specimen Transport Bags	Transport Bag, Specimen; Fisherbrand; Zippered; Polyethylene; Three-wall; With attached document compartment; Comply with OSHA and NCCLS guidelines; Large printed biohazard graphic; Size: 8 x 10 in.	Fisher Scientific	01-800-08	1 per cryovial box
Absorbent Pouch	6 Bay Aquipak, Absorbent Pak, Therapak	VWR	89170-936 (Therapak 10316)	1 per PBMC collection
Saf T Pak Leak Proof Bag and Tyvek Outer Envelope	Disposable 2-Part Secondary Pressure Vessel (95KPA), Medium; Inner Leak Proof Polybag 9.25 X 12in, Tyvek outer envelope, 7.5 X 9.5in	ESBE Scientific	STP-710	1 per PBMC collection
<b>FROZEN SHIPMENTS</b>				
Insulated Shipper	Shipper, Insulated; ThermoSafe; For Pharmaceutical Reagents; Economical; Lightweight; Reusable; Recyclable; Two-piece shippers with extra-thick insulating walls of expanded polystyrene; Dimensions (outside): 14-7/8 x 13-1/8 x 12-1/8 in.; Dimensions (inside): 11-3/4 x 10 x 9-1/8 in	Fisher Scientific	03-529-1	About two (2) 81 cell boxes fit per shipper with dry ice
<b>AMBIENT SHIPMENTS</b>				
Credo Cube Series 22 -248	Credo Cube™ Series 22, 2L, +15°C -+25°C, PCM Coolant, 53h Duration, 127.5m³; Dimensions (outside, mm): 276 X 250 X 241; 4.4kg	Fischer Scientific	NC1578810 (Pelican GH02APLAT CM22C-WT)	These boxes are re-usable and will be shipped back to the site

Biosample Collection Form [blood] (Appendix 8.1) and Biosample Collection Form [CSF] (Appendix 8.2) must be completed by the person collecting the biosample.

Patient Participant		
Visit	Timepoint	Suggested Collection Protocol
2	0 Month	2 RTT (10mL) → Serum 5 GTT (Heparin, 10mL) → PBMC 3 PTT (K <sub>2</sub> EDTA, 10mL) → Plasma (1X), DNA (2X) 2 PAXgene RNA (2.5mL) → RNA <b>Total Blood Tubes: 12</b> <b>Total Blood Volume: 105mL</b> CSF (up to 16mL) → optional procedure for patients only
3	4 Month	2 RTT (10mL) → Serum 1 PTT (K <sub>2</sub> EDTA, 10mL) → Plasma <b>Total Blood Tubes: 3</b> <b>Total Blood Volume: 30mL</b> CSF (up to 16mL) → optional procedure for patients only
4	8 Month	2 RTT (10mL) → Serum 1 PTT (K <sub>2</sub> EDTA, 10mL) → Plasma <b>Total Blood Tubes: 3</b> <b>Total Blood Volume: 30mL</b> CSF (up to 16mL) → optional procedure for patients only
5	12 Month	2 RTT (10mL) → Serum 1 PTT (K <sub>2</sub> EDTA, 10mL) → Plasma <b>Total Blood Tubes: 3</b> <b>Total Blood Volume: 30mL</b> CSF (up to 16mL) → optional procedure for patients only

Control Participant		
Visit	Timepoint	Suggested Collection Protocol
2	0 Months	2 RTT (10mL) → Serum 1 PTT (K <sub>2</sub> EDTA, 10mL) → Plasma 2 PAXgene RNA (2.5mL) → RNA <b>Total Blood Tubes: 5</b> <b>Total Blood Volume: 35mL</b>
3	8 Months	2 RTT (10mL) → Serum 1 PTT (K <sub>2</sub> EDTA, 10mL) → Plasma <b>Total Blood Tubes: 3</b> <b>Total Blood Volume: 30mL</b>



## 7.2. Recommended Order of Blood Draw Tubes

1. Red Top Tubes (Serum)
2. Green Top Heparin Tubes (PBMC)
3. Purple Top Tubes (Plasma, DNA)
4. PAXgene Tubes (RNA)

## 7.3. Managing Biosamples in the LORIS Database

The LORIS database (<https://cbigr.loris.ca/>), login using unique username and password) will be used to manage all samples collected through CAPTURE ALS. Personnel managing biosamples will need to request access to the Laboratory Information Management System (LIMS) module of LORIS (see LORIS manual). Samples will be collected, processed, stored, and shipped using the guidelines described below in sections 7.5 to 7.11. Processing of each sample type must be documented in LORIS. Detailed instructions and LORIS screenshots can be found in the LORIS manual. Paper processing forms may be used as an intermediary if data entry is not done directly in LORIS. The processing forms, specific to each sample type, can be found in the Appendix: Sample processing form - Serum (Appendix 8.3), Sample Processing Form - Plasma (Appendix 8.5), Sample Processing Form – CSF (Appendix 8.7). Sample Processing Form - PBMC (Appendix 8.4) and RNA processing form (Appendix 8.6) are included but may not be required by sites, as these samples will, in most cases, be processed by the C-BIG Repository. Any deviations to the sample processing protocols should be documented in the *comment section* of LORIS. Each sample will be identified with a label containing a QR code which will allow it to be scanned into LORIS (see section 7.4.4.2).

## 7.4. Generating and Printing Labels Overview

This section describes the processes to create labels for CAPTURE ALS biosamples (vacutainers and cryovials) at local sites, only. Labels generated at C-BIG will follow the existing LORIS formatting.

### 7.4.1. Collection Tube and Cryovial Labels

Each collection tube (vacutainer or 15mL tube) and cryovial label should contain the following information:

- **CAPTURE ALS Participant ID:** CAPTXXXXXXX
- **Vial #:** A unique number that should never be repeated within a participant
- **Sample type:** serum, plasma, CSF, DNA, RNA, PBMC
- **Visit Label:** 00M, 04M, 08M, 12M
- **Collection Date:** DD/MM/YYYY
- **QR code:** generated automatically from CAPT ID and Vial #

### 7.4.3. Creating Labels

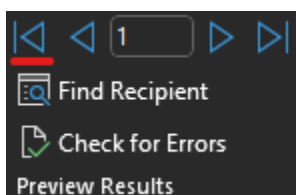
#### 7.4.3.1. Biosample Label Excel Template

- Use the provided biosample label template (Appendix 8.8) to generate labels for collection and local processing of biosamples.
  - This file contains four sheets, one for each visit (00M, 04M, 08M, 12M).
  - Each sheet contains all possible labels for a visit, including labels for vacutainers to collect whole blood or CSF, and labels for cryovials containing processed samples. Note: not all labels may be required for a particular visit. Consult the protocol and section 7.1 to determine what samples are collected at each visit.
  - Rows 2-15 contain vacutainer labels. The temporary labels contain the PSCID or CAPTURE ID (CAPTXXXXXX), unique vial #, sample type, visit label (XXM), collection date (DD/MM/YYYY) and barcode. The “NewCode” will be automatically generated from LORIS and will be Year000000 (where the year will be follow by a random number)
  - Rows 16-85 contain cryovial labels. The template will generate 15 serum labels, 10 plasma labels and 45 CSF labels. Labels contain the PSCID or CAPTURE ID (CAPTXXXXXX), unique vial #, sample type (serum, plasma, CSF), visit label (XXM), collection date (DD/MM/YYYY) and barcode. The “NewCode” will be automatically generated from LORIS and will be Year000000 (where the year will be follow by a random number)
- Prior to each participant visit, complete the required fields (Xs, DD/MM/YYYY) in the label template. Ensure that the sheet that corresponds to the correct visit (00M, 04M, 08M, 12M) is used. Never modify the unique vial #, as this number is used to generate a unique barcode. A barcode will be generated via the template, this barcode is temporary.
- Enter all the information into Loris as you are processing the biological samples. In Loris, the option of « generate barcode » will be available. Click on it and this new random code (YEAR000000) need to be use as your final code on your label on the cryovials. Copy this code into the column “NewCode” in the template file and generate/print now your labels.

#### 7.4.3.2. Generating labels

- Once the biosample label template has been completed with the participant information, Open Word.
- Click “Open” and open the label layout template provided: “*FISH-C6 Label Template CAPTURE ALS*”.
- Go to “Mailings” tab. Under the “Start Mail Merge”, select “Labels” and click “Cancel” in the Label Options pop up to activate the label template specified in the current Word file.
- Select “Select Recipients” and select “Use an Existing List” option.
- Select the excel file containing biosample label data.

- Select the correct visit point (00M, 04M etc.).
  - Select “Edit Recipient List” and select only the rows that you wish to print. E.g. Do not select PBMC rows if you are not collecting PBMCs at this visit.
  - Select “Insert Merge Field” and select all the options (PSCID, Vial #, Type, Visit Label, Collection Date, Barcode) one at a time by pressing “insert” after each option is selected.
  - Make sure the font of «PSCID» «Vial\_» «Type» «VisitLabel» «CollectionDate» is set to “Calibri (Body)” and font size to 4.
  - Add a space after each selection (e.g. «PSCID»<sub>space</sub>«Vial\_»).
  - Press enter to bring “«Barcode»” to a separate line and right click on “Toggle Field Codes”.
  - Replace MERGEFIELD with MERGEBarcode so that it reads MERGEBarcode Barcode Right click on MERGEBarcode Barcode to select the option “Edit Barcode”.
  - Under the “Barcode type” select QR Code.
  - On the bottom left, select “Advanced” and set scale to 28%.
  - Close by pressing OK.
  - Center the labels and select “Update Labels”.
  - Select “Preview Results”.
- Note: You may see incorrect number of labels or 1 label only as Word will shift to the last label hiding others.*
- Press on the icon shown in the image below underlined in red to shift back to the first label.



- Select “Finish & Merge”, “Print Documents” and then select “All” to print out the labels.
  - Print to a laser printer
- Note: Only print labels from a word file. Never print from a PDF as it will make QR code unreadable by the scanner. Load the label paper into the printer tray, do not use the manual feed as this may cause the labels to print out of the margins.*
- Test the QR codes using a barcode scanner to ensure that they can be read.

#### **7.4.3.3. Changing labels location**

A label sheet may have already been partially used. To start printing labels from a location other than the top left cell follow these steps:

- Finish creating labels as normal

- Cut the labels by highlighting the labels, right clicking the mouse and selecting the option “cut”
- Select the desired cell to paste the label selected above
- Right click the mouse and select “Paste” to move the label in selected cell

#### **7.4.4. Using Labels**

##### **7.4.4.1. Label placement**

Ensure that the label is placed **vertically** on the cryovial tube or vacutainer with the longest side of the label going up the length of the cryovial. DO NOT wrap the label around the tube, the QR code may not be readable.

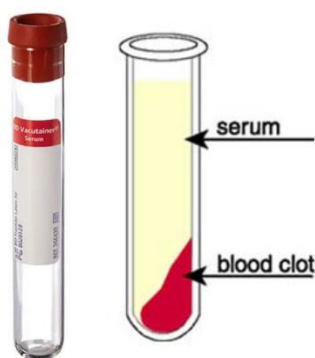
##### **7.4.4.2 Scanning labels into LORIS**

Each sample will be identified with a label containing a unique QR code. Following processing, the barcode scanner will be used to scan each QR code, entering the biosample into LORIS. This process links each biosample to a participant, visit and processing details. For full details see the LORIS manual. This step requires access to a computer.

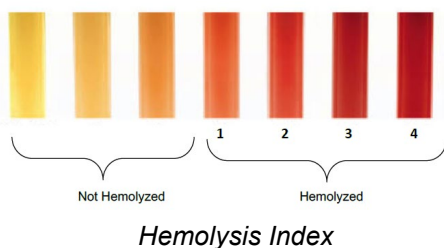
## 7.5. Whole Blood for Serum Isolation

Whole blood samples are collected at each visit for patient (MONTH 0, 4, 8, 12) and control participants (MONTH 0, 8). Samples are processed **locally** at each site for serum isolation. Two (2) tubes are suggested for this type of collection/visit.

### WHOLE BLOOD COLLECTION FOR SERUM



(BD Diagnostic, Cat# 366430)



Normal  
serum      Lipemic  
(milky)  
serum

### PREPARATION

Prepare two (2) 10mL red top tubes (RTT) by affixing labels. Prepare up to fifteen (15) 1.0mL cryovials by affixing labels.

### COLLECTION

- Completely fill two (2) 10mL RTT with blood.
- Complete the *Biosample Collection Form [blood]* (Appendix 8.1).
- Keep tube upright at room temperature for a minimum of 30min and maximum of 120 min to allow blood to clot.

### PROCESSING

- Serum isolation will be performed at the local site following SOP CBIG-02-003 titled “Serum Isolation from Whole Blood” (01May2020) as summarized below. Document serum processing on LORIS or by first completing the *serum processing form* (Appendix 8.3)
- Centrifuge at room temperature for 10 min at 2500Xg using a centrifuge with a swinging bucket.
- Indicate the degree of hemolysis (0-4) and whether the sample is lipemic (milky).
- Using a sterile transfer pipette, transfer supernatant (serum) to a new 15mL conical tube without disturbing the pellet.
- Using a micropipette or sterile transfer pipette, transfer 0.5mL aliquots into the 1.0mL cryovials, filling as many vials as possible.

### STORE FROZEN

- Store in a -80°C freezer until ready for shipment.
- Only if absolutely needed, aliquots can be stored at -20 °C for up to 1 week before transferring to -80 °C.

### SHIP

- Split the cryovials into two 81-cell freezer storage boxes, i.e., half of the samples in one box, half in the other. Each box should be shipped separately.
- For each participant, group samples by type and time point in the boxes.
- Bulk ship frozen samples on dry ice (see 7.11.4).

## 7.6. Whole Blood for PBMC Isolation

Whole blood samples are collected from patient participants at a single time-point (MONTH 0 suggested) and shipped at room temperature to C-BIG for PBMC isolation. Five (5) tubes are suggested for this type of collection. No aliquoting is required.

### WHOLE BLOOD COLLECTION FOR PBMC



(BD Diagnostic, Cat# 366480)

#### PURPOSE OF COLLECTION

PBMC isolation at C-BIG.

#### PREPARATION

Prepare five (5) 10mL green top sodium heparin tubes (GTT) by affixing labels.

Ensure the tubes are at room temperature (15-25°C).

**Warning:** Green vacutainers require careful handling, they are made of glass.

#### COLLECTION

- Completely fill five (5) 10mL green top sodium heparin tubes with blood.
- Gently invert tube 8-10 times immediately after blood draw.
- Complete the *Biosample Collection Form [blood]* (Appendix 8.1).

#### SHIP

- Vacutainers should be kept at room temperature (15-25°C) and shipped immediately.
- Ship priority overnight at ambient temperature in Credo Cube to C-BIG (see 7.11.5).

#### PROCESSING

PBMC isolation will be performed by C-BIG following SOP CBIG-02-002 titled "PBMC Isolation from Whole Blood (Leucosep Method) (30Apr2020). Only if PBMC processing is done locally, does the *PBMC processing form* (Appendix 8.4) need to be completed on LORIS.

### 7.7. Whole Blood for DNA Extraction

Whole blood samples are collected from patient participants at MONTH 0 and shipped to C-BIG for DNA extraction. Two (2) tubes are suggested for this type of collection. No aliquoting is required.

#### WHOLE BLOOD COLLECTION FOR DNA



(BD Diagnostic, Cat# 366643)

#### PURPOSE OF COLLECTION

DNA extraction at C-BIG.

#### PREPARATION

Prepare two (2) 10mL purple top K<sub>2</sub>EDTA tubes (PTT) by affixing labels.

#### COLLECTION

- Completely fill two (2) 10mL purple top K<sub>2</sub>EDTA tubes with blood.
- Complete the *Biosample Collection Form [blood]* (Appendix 8.1).

#### STORE FROZEN

- Store vertically in a -80°C freezer until ready for shipment.
- Only if absolutely needed, vacutainer can be stored at -20 °C for 2 days before transferring to -80 °C.
- **Warning:** Tubes will expand during freezing, avoid storing them in a confined rack (e.g. Styrofoam rack).

#### SHIP

- Place the frozen tube(s) in a biohazard bag.
- DNA tubes from a single participant should be shipped separately from one another.
- Bulk ship frozen samples on dry ice (see 7.11.4).

#### PROCESSING

DNA extraction will be performed by C-BIG following SOP CBIG-02-004, titled “DNA Extraction from Whole Blood (Accelerated Method)” (01May2020).



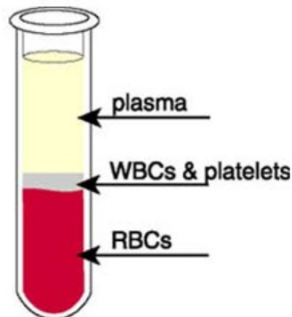
## 7.8. Whole Blood for Plasma Isolation

Whole blood samples are collected at each visit for patient (MONTH 0, 4, 8, 12) and control participants (MONTH 0, 8). Samples are processed **locally** at each site for plasma isolation. One (1) tube is suggested for this type of collection/visit.

### PLASMA COLLECTION



(BD Diagnostic, Cat# 366643)



*Plasma is the top layer. White blood cells and platelets are in a whitish layer called the “buffy coat”, just under the plasma and above the red blood cells.*

### PREPARATION

Prepare one (1) 10mL purple top K<sub>2</sub>EDTA tube (PTT) by affixing a label.

Prepare up to ten (10) 1.0mL cryovials by affixing labels.

### COLLECTION

- Completely fill one (1) purple top K<sub>2</sub>EDTA tube with blood.
- Store purple top tube upright at room temperature away from direct light for a minimum of 30 min to allow blood to sediment.
- Complete the *Biosample Collection Form [blood]* (Appendix 8.1).

### PROCESSING

- Plasma isolation will be performed at the local site following SOP CBIG-02-011 titled “Plasma Isolation from Whole Blood” (02Oct2020) as summarized below. Document plasma processing on LORIS or by first completing the *plasma processing form* (Appendix 8.5).
- Centrifuge at 4°C for 12 min at 250Xg using a centrifuge with a swinging bucket.
- Using a sterile transfer pipette, quickly transfer supernatant (plasma) to a new 15mL polypropylene conical tube.
- **Warning:** Do not disturb the buffy coat.
- Using a micropipette or sterile transfer pipette, transfer 0.5mL aliquots into the 1.0mL cryovials, filling as many vials as possible.

### STORE FROZEN

- Plasma samples can be transferred directly to -80°C freezer until shipment.
- Only if absolutely needed, aliquots can be stored at -20 °C for 2 days before transferring to -80 °C.



	<p><b>SHIP</b></p> <ul style="list-style-type: none"><li>• Split the cryovials into two 81-cell freezer storage boxes, i.e., half of the samples in one box, half in the other. Each box should be shipped separately.</li><li>• For each participant, group samples by type and time point in the boxes.</li><li>• Bulk ship frozen samples on dry ice (see 7.11.4).</li></ul>
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### 7.9. Whole Blood for RNA Extraction

Whole blood samples are collected from patient and controls participants at MONTH 0 and shipped to C-BIG for RNA storage. Two (2) tubes are suggested for this type of collection. No aliquoting is required.

#### WHOLE BLOOD COLLECTION FOR RNA



(BD Diagnostic, Cat# 762165)

#### PURPOSE OF COLLECTION

RNA storage at C-BIG.

#### PREPARATION

Prepare two (2) 2.5mL PAXgene RNA tubes by affixing labels.

#### COLLECTION

- Completely fill two (2) 2.5mL PAXgene RNA tubes with blood.
- Gently invert tube 8-10 times immediately after blood draw.
- Complete the *Biosample Collection Form [blood]* (Appendix 8.1).

#### STORE FROZEN

- Follow protocol “CAPTURE ALS RNA Processing (07Mar2022)” as summarized below and found in Appendix 8.6. This “processing” does not need to be documented in LORIS.
- Keep tubes upright at room temperature for a minimum of 2 hours and a maximum of 72hours to allow blood to clot.
- Transfer tubes to -20°C for at least 24hours before transferring to -80°C freezer until ready for shipment.

#### SHIP

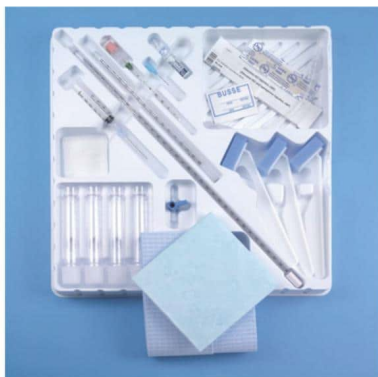
- Place the frozen tube(s) in a biohazard bag.
- RNA tubes from a single participant should be shipped separately from one another.
- Bulk ship frozen samples on dry ice (see 7.11.4).

#### PROCESSING / LONG-TERM STORAGE

RNA tubes will be stored by C-BIG at -80°C for future RNA extraction.

## 7.10. Cerebrospinal Fluid (CSF) Collection (optional procedure)

### CSF COLLECTION



*Local site's preferred lumbar puncture tray*

### PREPARATION

Prepare lumbar puncture tray and additional supplies and two (2) 15mL polypropylene tubes by affixing labels.

Prepare up to forty-five (45) 1.0mL cryovials by affixing labels.

**Warning:** *Coagulation and platelets tests must be reviewed by a MD within 4 weeks of performing a lumbar puncture.*

### COLLECTION

- A licensed practitioner will collect **up to 16mL** of CSF in accordance with local site SOPs.
- CSF should be collected in polypropylene tubes due to their low protein binding potential, no additives should be used.
- Immediately after collection, keep the CSF at 4°C or on ice until ready to process.
- Complete the *Biosample Collection Form [CSF]* (Appendix 8.2).

### PROCESSING

- Send 1-2ml of collected CSF to local laboratory for cell count, protein and glucose analysis, follow local laboratory guidelines.
- CSF processing will be performed at the local site following SOP *CBIG-02-008 titled "CSF Processing" (01May2020)* as summarized below. Document CSF processing on LORIS or by first completing the *CSF processing form* (Appendix 8.7).
- Process the sample promptly.
- Using a sterile transfer pipette, transfer the CSF to a new 15mL polypropylene tube.
- Centrifuge at room temperature for 10min at 2000Xg using a centrifuge with a swinging bucket.
- Using a sterile transfer pipette, transfer the supernatant (CSF) to a new 15mL polypropylene tube, do not disturb the pellet.
- Using a micropipette or sterile transfer pipette, transfer 0.5mL aliquots into the 1.0mL cryovials, filling as many vials as possible.

	<p><b>STORE FROZEN</b></p> <ul style="list-style-type: none"> <li>• Store cryovials directly in a -80°C freezer until ready for shipment. CSF cannot be stored at -20°C.</li> </ul> <p><b>SHIP</b></p> <ul style="list-style-type: none"> <li>• Split the cryovials into two 81-cell freezer storage boxes, i.e., half of the samples in one box, half in the other. Each box should be shipped separately.</li> <li>• For each participant, group samples by type and time point in the boxes.</li> <li>• Bulk ship frozen samples on dry ice (see 7.11.4).</li> </ul>
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## 7.11. PACKAGING AND SHIPPING INSTRUCTIONS

### 7.11.1. Shipping to C-BIG Repository

- Samples will be shipped either frozen on dry ice or at ambient temperature in a credo cube.
- Frozen samples will be shipped in two batches to prevent the loss of all samples from a participant in the event a shipment is lost or thawed prior to delivery.
- Samples will be shipped on Monday to Wednesday for delivery on Tuesday to Thursday. Avoid shipping on Friday to prevent sample delivery on a weekend.
- Prior to shipment of samples, confirmation of shipment date, via telephone or email, shall be made with the C-BIG coordinator.
- To schedule a shipment, visit <https://www.fedex.com/en-ca/shipping.html>
- Sites will receive a sample shipment confirmation from C-BIG after samples arrive with instructions when to ship the second box.

### 7.11.2. Shipping Address

**Mailing Address:** Attention: Marie-Noelle Boivin  
C-BIG Repository  
Montreal Neurological Institute  
3801 University Street  
Room NWB150  
Montreal, Quebec  
H3A 2B4

**Phone Number:** 514-398-8439

**Email:** [cbig.mni@mcgill.ca](mailto:cbig.mni@mcgill.ca)

### 7.11.3. C-BIG Hours of Operation and Holidays

Monday to Friday 7:30 am to 4:30 pm Eastern Time

C-BIG lab coordinators will be available to receive shipments from 7:45AM to 3:00PM.

If a shipment at C-BIG is anticipated outside of these hours, an arrangement **MUST** be made with the C-BIG coordinator at the contact information above.

### Holiday Observations

Based on previous years, FedEx observes the following holidays. These dates are subject to change by FedEx.

It is ***strongly recommended*** that you verify FedEx and C-BIG schedules prior to any holiday shipments to Montreal.

#### Avoid collecting specimens for shipment on the day before an observed holiday

New Years Day	January 1
Good Friday	Date varies yearly
Easter Monday	Date varies yearly
Victoria Day	The second to last Monday in May
St. Jean Baptiste Day	June 24
Canada Day	July 1
Labor Day	1 <sup>st</sup> Monday in September
Thanksgiving	2 <sup>nd</sup> Monday in October
Christmas Day	December 25*
Boxing Day	December 26*

\*C-BIG will inform Program Manager of C-BIG Christmas Holiday closure dates.

#### **7.11.4. Frozen Sample Shipments**

The following samples are shipped frozen on dry ice to C-BIG:

1. Serum, plasma, and CSF cryovials
2. Purple Top DNA Collection Vacutainer(s)
3. PAXgene RNA Collection Vacutainer(s)

##### **7.11.4.1. Frozen Sample Shipping Supplies**

- Biohazard Specimen Transport Bags

- Absorbent Sheet(s)
- Dry Ice Label(s)
- UN 3373 Biological Substance Category B Label(s)
- Insulated Shipping Container(s)
- Dry Ice
- Packing Tape

#### 7.11.4.2. Frozen Sample Packing Instructions

- Order dry ice from your preferred supplier. 10lbs of dry ice is required for the suggested insulated Styrofoam shipping container.
- Complete the packing slip (see Appendix 8.9) and place it in the shipping container. Retain a copy for site records. If DNA or RNA tubes are being sent, include a copy of the Biosample Collection Form [Blood] (Appendix 8.1) showing the collection details. This will facilitate processing at C-BIG.
- Cryovials: Place each 81-cell sample cryobox in a biohazard specimen transport bag with an absorbent sheet. **Do not tape the 81-cell sample box.**
- Whole Tubes: Place frozen whole blood tube(s) in a biohazard bag with an absorbent sheet.
- Place up to two (2) biohazard specimen transport bags each containing a single 81-cell box in an insulated Styrofoam shipping container. A third biohazard bag containing whole blood tubes may be added to the same shipping container.
- Surround samples on all sides (bottom, top, sides) with dry ice (10lbs). **Do not tape the insulated Styrofoam shipping container.**
- Seal the cardboard shipping box with standard packing tape.

#### 7.11.4.3. Frozen Sample Shipping Instructions

- Affix the 'Substance B' shipping label to the outside of the cardboard shipping box.
- Complete the 'Dry Ice' shipping label and affix to the outside of the shipping box.

Enter the number of kg of dry ice placed in the cooler.

Enter your shipping name and address.

shippers declaration not required

part B is required

dry ice amount must be in kilograms

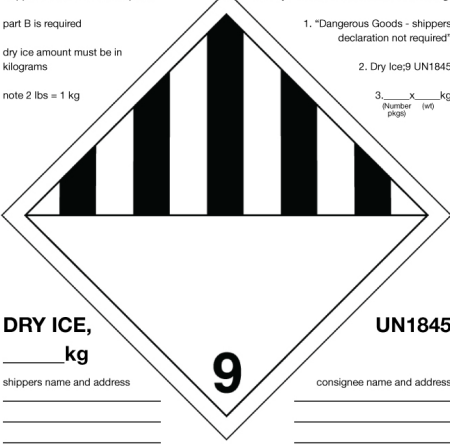
note 2 lbs = 1 kg

airwaybills/airbills must have the following:

1. "Dangerous Goods - shippers declaration not required"

2. Dry Ice;9 UN1845

3.  $\frac{\text{Number}}{\text{pkg}}$   $\times$   $\frac{\text{wt}}{\text{kg}}$



**DRY ICE,**  
\_\_\_\_ kg

shippers name and address

\_\_\_\_\_  
\_\_\_\_\_

consignee name and address

\_\_\_\_\_  
\_\_\_\_\_

**UN1845**

Enter '1' on *Number pkgs* line and weight in kg of entire box on *wt* line.

- Complete the FedEx Intra-Canada Air waybill with the CAPTURE ALS account number. CAPTURE ALS will be charged for the shipment.
  - Section 1/From: Complete Date, Sender's Name & Shipping Address
  - Section 2/Internal Reference: CAPTURE ALS, ZAEXK
  - Section 3/To: Enter Recipient's Name & Address
    - Recipient Name: C-BIG (M.N. Boivin)
    - Phone: 514-398-8439
    - Company: CBIG MNI -MCGILL U
    - Address (line 1): Neuro Immunology
    - Address (line 2): 3801 Rue University, Room NWB150
    - City: Montreal
    - Province: QC
    - Postal Code: H3A 2B4
  - Section 4/Shipment Information: Complete number packages, total weight and value.
  - Section 5a/Express Package Services: Check "FedEx Priority Overnight"
  - Section 6/Packaging: Check "Other", write "Insulated Shipper"
  - Section 7/Special Handling:
    - Check "YES - Shippers Declaration Not Required"
    - Check "Dry Ice" and enter the total weight of dry ice (10lbs).
  - Section 8/Payment: Check "Third Party", FedEx Acct No. 111266182

**FedEx Intra-Canada Air Waybill**  
Lettre de transport aérien intérieure

Not all services and options are available to all destinations.  
Certains services et options ne sont pas disponibles pour toutes les destinations.

1 From / Please print and press firmly / Expéditeur / Écrivez en caractères d'imprimerie. Appuyez fermement sur l'empreinte.

Sender's Name / Nom de l'expéditeur: CAPTURE ALS  
Address / Adresse: 3801 RUE UNIVERSITY  
City / Ville: MONTREAL  
Province / Province: QC  
Postal Code / Code postal: H3A 2B4

2 To / Destinataire  
Recipient's Name / Nom du destinataire: CBIG (M.N. Boivin)  
Address / Adresse: NEURO IMMUNOLOGY  
City / Ville: MONTREAL  
Province / Province: QC  
Postal Code / Code postal: H3A 2B4

3 Shipment Information / Informations sur l'envoi  
Total Packages / Nombre total de colis: 1  
Total Weight / Poids total: 10 kg  
Total Declared Value / Valeur déclarée: \$

4a Express Package Service / Service colis express  
FedEx First Overnight  
☒ FedEx Priority Overnight  
FedEx Standard Overnight

4b Express Freight Service / Service fret express  
FedEx 10 Day Freight

5 Packaging / Emballage  
☒ FedEx Envelope / Enveloppe FedEx  
☐ FedEx Pak / Pak FedEx  
☐ FedEx Box / Boîte FedEx  
☒ Insulated Shipper

6 Special Handling and Delivery Signature Options / Options de manutention spéciale et de signature de livraison  
☒ HOLD at FedEx Location / RETENIR à la succursale FedEx  
☐ SATURDAY Delivery / Livraison le SAMEDI  
☐ Direct Signature / Signature directe  
☐ Indirect Signature / Signature indirecte

7 Payment / Paiement  
Bill transportation charges to / Facturer le transport à:  
Sender / Expéditeur  
Recipient / Destinataire  
Third Party / Tierce partie  
Credit Card / Carte de crédit  
Cash/Check / Argent liquide/Chèque

8 Required Signature / Signature requise  
Sender's Signature / Signature de l'expéditeur  
Recipient's Signature / Signature du destinataire  
Third Party Signature / Signature de la tierce partie  
Credit Card Signature / Signature de la carte de crédit

FedEx Tracking Number / Numéro de suivi de FedEx: 8168 6991 1269  
Form ID No. / N° de formulaire: 0450

- Schedule a pick-up time and drop off the box at the pick-up location
- Email the biobank at [cbig.mni@mcgill.ca](mailto:cbig.mni@mcgill.ca) or call the Biobank at 514-398-8439 to let them know which samples are being sent as well as the tracking number.



#### **7.11.5. Ambient Sample Shipments (Credo)**

Green Top Blood Collection Tubes (GTTs; for PBMC isolation) are shipped at controlled ambient temperature to C-BIG in a credo cube.

##### **7.11.5.1. Ambient Sample Shipping Supplies**

1. GTTs for PBMC Isolation
2. Elastic
3. Parafilm
4. Biohazard specimen transport bag
5. Absorbent Pouch
6. Safe T Pak Leak proof bag and Tvek outer envelope
7. Credo Box
8. Packing Tape

##### **7.11.5.2. Ambient Sample Packing Instructions**


- The GTTs should be kept at room temperature and sent on the same day as collection.
- A single credo cube can contain PBMC collections from two (2) participants collected on the same day. The following steps should be followed for each participant.
- Complete the packing slip (Appendix 8.9) and place it in the shipping container. Retain a copy for site records. Include a copy of the Biosample Collection Form [Blood] (Appendix 8.1) to facilitate sample processing at C-BIG.
- Place parafilm on the lids of the GTTs that have been filled.
- Place up to 6 GTTs, from a single participant, inside the absorbent Aquipak pouch.
- Roll the absorbent pouch and attach it with an elastic.
- Place the absorbent protected vacutainers in the biohazard bag.
- Place the biohazard bag containing vacutainers in the Saf T Pak leak-proof poly bag (part #1 of STP-710) and seal.
- Place the leak-proof poly bag in the Tyvek outer envelope and seal (part #2 of STP-710) and seal.
- Repeat steps above as necessary for additional GTTs.
- Place the biohazard envelope in the Credo Cube.
- Seal the cardboard shipping box with standard packing tape.

##### **7.11.5.3. Ambient Sample Shipping Instructions**

- Affix the 'Substance B' shipping label to the outside of the cardboard shipping box.
- Complete the FedEx Intra-Canada Air waybill with the CAPTURE ALS account number. CAPTURE ALS will be charged for the shipment.
  - Section 1/From: Complete Date, Sender's Name & Shipping Address



- Section 2/Internal Reference: CAPTURE ALS, ZAEXK
- Section 3/To: Enter Recipient's Name & Address
  - Recipient Name: C-BIG (M.N. Boivin)
  - Phone: 514-398-8439
  - Company: CBIG MNI -MCGILL U
  - Address (line 1): Neuro Immunology
  - Address (line 2): 3801 Rue University, Room NWB150
  - City: Montreal
  - Province: QC
  - Postal Code: H3A 2B4
- Section 4/Shipment Information: Complete number packages, total weight and value.
- Section 5a/Express Package Services: Check "FedEx Priority Overnight"
- Section 6/Packaging: Check "Other", write "Insulated Shipper"
- Section 7/Special Handling:
  - Check "NO"
- Section 8/Payment: Check "Third Party", FedEx Acct No. 111266182



## Intra-Canada Air Waybill Lettre de transport aérien intérieure

Net air services and options available to all destinations.  
Certains services et options ne sont pas disponibles pour toutes les destinations.

**Sender's Copy**  
Copie de l'expéditeur

**Flight** **DESTIN**  
N° de vol

**Origin** **ORIGIN**  
N° de vol

**DATE**  
Date

**TIME**  
Heure

**Form 22**  
Form 22

**Form 22**  
Form 22

**1 From Place and person filled, Expéditeur/Écrire en caractères d'imprimerie. Apposer fermement SVP**  
Sender's FedEx Account Number  
N° de compte FedEx de l'expéditeur

**2 To/Destination**  
CAPTULE ALS, ZAEKK

**Sender's Name**  
Nom de l'expéditeur

**Company**  
Société de l'expéditeur

**Address**  
Adresse

**City**  
Ville

**Postal Code**  
Code postal

**Province**  
Province

**Phone**  
Téléphone

**Fax**  
Téléfax

**Form 22**  
Form 22

**Form 22**  
Form 22

**3 Shipment Information/Informations sur l'envoi**

**Total Packages**  
Nombre total de colis

**Total Weight**  
Poids total

**Total Declared Value**  
Valeur déclarée des colis

**4a Express Package Service/Service colis express**

☐ FedEx First Overnight ☒ FedEx Priority Overnight ☐ FedEx Standard Overnight

☐ FedEx 2Day ☐ FedEx Economy ☐ FedEx Next Business Day

**4b Express Freight Service/Service fret express**

☐ FedEx 1 Day Freight

**5 Packaging/Emballage**

☐ FedEx Envelope ☐ FedEx Pak ☐ FedEx Box ☐ FedEx Tube ☐ FedEx Tube ☒ Insulated Shipper

**6 Special Handling and Delivery Signature Options**  
Options de manutention spéciale et de signature de livraison

☐ HOLD at FedEx Location ☐ SATURDAY Delivery / Livraison le SAMEDI

☐ RETAIN at the origin ☐ FedEx Signature ☐ FedEx Signature ☐ FedEx Signature

**7 Payment / Paiement**

☐ Cash/Check ☐ Credit Card

**8 Required Signature/Signature requise**

☐ Sender's Signature ☐ Recipient's Signature

**9 Insurance**

☐ Insured ☐ Uninsured

**10 Tracking Number**  
N° de suivi

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**84 Tracking Number**  
N° de suivi

**85 Tracking Number**  
N° de suivi

**8**

- Schedule a pick-up time and drop off the box at the pick-up location
- Email the biobank at [cbig.mni@mcgill.ca](mailto:cbig.mni@mcgill.ca) or call the Biobank at 514-398-8439 to let them know which samples are being sent as well as the tracking number.

**7.11.5.4. Credo Return Shipping Instructions**

- C-BIG will return credo boxes to each site using FedEx Ground and Acct No. 111266182.
- Ensure that “Collect” is checked off on the Ground waybill when items are shipped to or from the University of Alberta.

## **8. APPENDICES**

**Appendix 8.1: CAPTURE ALS Biosample Collection Form [Blood]**

**Appendix 8.2: CAPTURE ALS Biosample Collection Form [CSF]**

**Appendix 8.3: Sample Processing Form - Serum**

**Appendix 8.4: Sample Processing Form - PBMC via Leucosep**

**Appendix 8.5: Sample Processing Form - Plasma**

**Appendix 8.6: Sample Processing Form - RNA**

**Appendix 8.7: Sample Processing Form - CSF**

**Appendix 8.8: Biosample Labels Template**

**Appendix 8.9: CAPTURE ALS Packing Slip**

## Appendix 8.1: CAPTURE ALS Biosample Collection Form [Blood]

Found in [Biosample Collection Form [Blood]] (04Mar2025) CAPTURE ALS



CAPTURE ALS

### Biosample Collection Form [Blood]

SECTION 1. PARTICIPANT						
CONTROL <input type="checkbox"/> PATIENT <input type="checkbox"/>			CAPTURE-ALS ID: CAPT_____			
SECTION 2. BASIC DEMOGRAPHIC INFORMATION						
SEX:      MALE <input type="checkbox"/> FEMALE <input type="checkbox"/>			Date of birth:      MM / YYYY			
SECTION 3. STUDY INFORMATION						
Study Name: CAPTURE-ALS						
Visit Month:      0 <input type="checkbox"/> 4 <input type="checkbox"/> 8 <input type="checkbox"/> 12 <input type="checkbox"/>						
Site:      Edmonton <input type="checkbox"/> Montreal <input type="checkbox"/> CHUM <input type="checkbox"/> Toronto <input type="checkbox"/> Quebec City <input type="checkbox"/>						
Current Diagnosis:      ALS <input type="checkbox"/> ALS-related disease <input type="checkbox"/> Healthy Control <input type="checkbox"/> Other <input type="checkbox"/> : _____						
SECTION 4. BLOOD DRAW						
<p>[Patient Participant]</p> <p><b>0 Month (Total tubes:12)</b> 2 RIT (Serum), 5 GTT (PBMC), 3 PTT (Plasma (1), DNA (2)), 2 PAXgene (RNA)</p> <p><b>4, 8, 12 Month (Total tubes :3)</b> 2 RIT (Serum), 1 PTT (Plasma)</p>		VACUTAINERS				
		Purpose	Color	Type	# Requested	# Drawn
<p>[Healthy Control Participant]</p> <p><b>0 Month (Total tubes: 5 / visit)</b> 2 RIT (Serum), 1 PTT (Plasma), 2 PAXgene (RNA)</p> <p><b>8 Month (Total tubes : 3 / visit)</b> 2 RIT (Serum), 1 PTT (Plasma)</p>		Serum	Red	No Additive	2	
		PBMC	Green	Heparin	5	
		Plasma	Purple	K <sub>2</sub> EDTA	1	
		DNA	Purple	K <sub>2</sub> EDTA	2	
		RNA		PAXgene	2	
Recommended Order of Blood Draw: RIT, GTT, PTT, PAXgene						
Drawn by:		Draw Date:		Draw Time:		
		DD / MM / YYYY		hh / mm		

## Appendix 8.2: CAPTURE ALS Biosample Collection Form [CSF]

Found in [Biosample Collection Form [CSF] (04Mar2025) CAPTURE ALS]



CAPTURE ALS

### Biosample Collection Form [CSF]

SECTION 1. PARTICIPANT				
PATIENT <input type="checkbox"/>			CAPTURE-ALS ID: CAPT_____	
SECTION 2. BASIC DEMOGRAPHIC INFORMATION				
SEX:      MALE <input type="checkbox"/> FEMALE <input type="checkbox"/>			Date of birth:      MM / YYYY	
SECTION 3. STUDY INFORMATION				
Study Name: CAPTURE-ALS				
Visit Month:      0 <input type="checkbox"/> 4 <input type="checkbox"/> 8 <input type="checkbox"/> 12 <input type="checkbox"/>				
Site:    Edmonton <input type="checkbox"/> Montreal <input type="checkbox"/> CHUM <input type="checkbox"/> Toronto <input type="checkbox"/> Quebec City <input type="checkbox"/>				
Current Diagnosis:    ALS <input type="checkbox"/> ALS-related disease <input type="checkbox"/> Healthy Control <input type="checkbox"/> Other <input type="checkbox"/> : _____				
SECTION 4. CSF DRAW				
<input type="checkbox"/> PT/PTT/INR/CBC levels verified by a physician prior to LP				
A licensed practitioner will collect up to 16mL of CSF.		VACCUTAINERS		
Must be collected in polypropylene tubes No additives used.		Purpose	Type	# Requested
1 tube (1-2 mL) goes to local lab for analysis of cell count, protein, glucose.		CSF	Polypropylene tubes	2
Immediately after collection, keep the CSF at 4°C				
LP performed by:		Draw Date:		Draw Time:
NAME      /      SIGNATURE		DD / MM / YYYY		hh / mm
<input type="checkbox"/> 1 tube of CSF (1-2 mL) sent to local lab for analysis				
<input type="checkbox"/> CSF collected for CAPTURE-ALS (volume of CSF collected: _____ mL)				
Note (did the participant experience any adverse event, etc.):				

### Appendix 8.3: Sample Processing Form - Serum

Found in [Sample Processing Form – Serum (7Mar2022) CAPTURE ALS]



Serum Isolation from Whole Blood  
Sample Processing Form: Serum  
Last updated: 7March2022

SERUM PROCESSING				SOP Employed: CBIG-02-003	
Processed by: _____			CAPT ID: _____		
Time start processing: _____			Date: ____/____/____		
VACUTAINERS/TUBES			QUANTITY		
RECEIVED	TYPE	RECEIVED	ALIQUOTS		
_____	_____	_____ mL	_____ x 500µL		
Issues	<input type="checkbox"/> Milky serum	<input type="checkbox"/> Hemolyzed, if checked, score (0-4)	<input type="checkbox"/> Expired tubes Date of expiry: ____/____/____		
RECEIVED	TYPE	RECEIVED	ALIQUOTS		
_____	_____	_____ mL	_____ x 500µL		
Issues	<input type="checkbox"/> Milky Serum	<input type="checkbox"/> Hemolyzed, if checked, score (0-4)	<input type="checkbox"/> Expired tubes Date of expiry: ____/____/____		
STEPS:					
<input type="checkbox"/> 1. Keep tube upright at room temperature for a minimum of 30min and a maximum of 120 min to allow blood to clot					
<input type="checkbox"/> 2. Centrifuge 10 minutes at 2500Xg Start: ____:____ End: ____:____					
<input type="checkbox"/> 3. Transfer SN to a new 15 mL Falcon tube					
<input type="checkbox"/> 4. Prepare aliquots (500 µL)					
<input type="checkbox"/> 5. Store at -80 °C (until ship to C-BIG) Date: ____/____/____					
REAGENTS TRACKING					
	Lot #	Expiry Date	Opened	Finished	
Vacutainers	_____	____/____/____	N/App	N/App	

Notes:

## Appendix 8.4: Sample Processing Form - PBMC via Leucosep

Found in [Sample Processing Form – PBMC via Leucosep (7Mar2022) CAPTURE ALS]



PBMC Isolation from Whole Blood (Leucosep Method)  
Sample Processing Form: PBMC via Leucosep  
Last updated: 7March2022

PBMC PROCESSING				SOP Employed: CBIG-02-002	
Processed by: _____			CAPT ID: _____		
Time start processing: ____:____			Date: ____/____/____		
VACCUTAINERS/TUBES		QUANTITY			
RECEIVED	TYPE	RECEIVED			
____	____	____ mL			
<b>Issues</b> <input type="checkbox"/> Red Pellet (RP) <input type="checkbox"/> Clotted <input type="checkbox"/> Expired tubes Date of expiry: ____/____/____					
<b>STEPS:</b>					
<input type="checkbox"/> 1. Distribute 15 mL of Ficoll into the Leucosep Tube					
<input type="checkbox"/> 2. Centrifuge for 30 seconds at 1000Xg					
<input type="checkbox"/> 3. Distribute the blood from the vacutainers and dilute 1:2 with PBS into the Leucosep Tube (up to 35 mL)					
<input type="checkbox"/> 4. Centrifuge for 10 minutes at 1000Xg, with NO break				Start: ____:____	End: ____:____
<input type="checkbox"/> 5. Collect PBMC by pouring into a new 50 mL tube and complete suspension to 50 mL with PBS					
<input type="checkbox"/> 6. Centrifuge for 10 minutes at 250Xg				Start: ____:____	End: ____:____
<input type="checkbox"/> 7. Discard supernatant, loosen pellet, add 10 mL of PBS					
<input type="checkbox"/> 8. Count cell					
<input type="checkbox"/> 9. Take cell if needed for trizol and complete suspension to 50 mL with PBS					
<input type="checkbox"/> 10. Centrifuge for 10 minutes at 250Xg				Start: ____:____	End: ____:____
<input type="checkbox"/> 11. Discard supernatant and loosen cell pellet					
<input type="checkbox"/> 12. Add solution A (HI HuAB serum)					
<input type="checkbox"/> 13. Add solution B (HI HuAB serum + DMSO)					
<input type="checkbox"/> Prepare aliquots		Date: ____/____/____			
<input type="checkbox"/> Store at -80°C		Date: ____/____/____			
<input type="checkbox"/> Store in liquid nitrogen		Date: ____/____/____			
PBMC COUNTING				SOP Employed: CBIG-03-001	
Dilution factor :2		Cell suspension volume:10.00mL			
(already take into account the dilution factor)		Countess FL II		Count #1	____ x10 <sup>6</sup> cells/ mL
				Count #2	____ x 10 <sup>6</sup> cells/mL
SUM OF COUNTS: Count #1 + Count #2		CELL COUNTING MEAN: Sum of Count #1 and Count #2 / 2		____	
TOTAL CELLS ISOLATED : Cell Counting Mean x Cell Suspension Volume (mL)					
Coefficient of Variance (% C.V.) (need to be under 5%)	Live: ____	Live: ____	Total PBMC	____ x10 <sup>6</sup>	
	Dead: ____	Dead: ____			
				<input type="checkbox"/> If trizol done take: ____ x 10 <sup>6</sup> cells	
				Final PBMC count (after trizol) ____ x 10 <sup>6</sup> cells	

Adapted from C-BIG PBMC via Leucosep Sample Processing Form, Dated: April 22, 2020






PBMC Isolation from Whole Blood (Leucosep Method)  
Sample Processing Form: PBMC via Leucosep  
Last updated: 7March2022

PBMC FREEZING				
FREEZING SOLUTION B PREPATION (mL) HuAB serum + 20% DMSO		HuAB: _____ mL DMSO: _____ mL	NUMBER OF VIALS	—
TOTAL FREEZING SOLUTION VOLUME USED (mL) 50% Solution A (HuAB serum) + 50% Solution B Total Volume = 1.00mL x Number of Vials		_____ mL	CELLS PER VIAL Total Cells Isolated / Number of Vials **SHOULD NOT EXCEED 21.50 x10 <sup>6</sup>	_____ x10 <sup>6</sup>
QUANTITY - ALIQUOTS				
_____ X vials at _____ x 10 <sup>6</sup> cells / mL in each vial				
REAGENTS TRACKING				
	Lot #	Expiry Date	Opened	Finished
Leucosep	_____	_____/____/____	N/App	N/App
PBS	_____	_____/____/____		
Ficoll	_____	_____/____/____		
HI HuAB serum	_____	_____/____/____		
DMSO	_____	- / -		
Vacutainers	_____	_____/____/____	N/App	N/App
Notes:				



## Appendix 8.5: Sample Processing Form - Plasma

Found in [Sample Processing Form – Plasma (7Mar2022) CAPTURE ALS]

		Plasma Isolation from Whole Blood Sample Processing Form: Plasma Last updated: 7March2022	
<b>PLASMA PROCESSING</b>		<b>SOP Employed: CBIG-02-011</b>	
Processed by: _____		CAPT ID: _____	
Time start processing: ____:____		Date: ____/____/____	
<b>VACUTAINERS/TUBES</b>		<b>QUANTITY</b>	
<b>RECEIVED</b>	<b>TYPE</b>	<b>RECEIVED</b>	
____	____	____mL	
<b>Concerns</b> <input type="checkbox"/> Expired tubes    Date of expiry: ____/____/____			
<b>STEPS</b>			
<input type="checkbox"/> 1. Store PTT in the dark and upright for 30 min		Start: ____:____	End: ____:____
<input type="checkbox"/> 2. Centrifuge for 12 minutes at 250Xg at 4°C		Start: ____:____	End: ____:____
<input type="checkbox"/> 3. Transfer supernatant into a new conical tube			
<input type="checkbox"/> 4. Prepare aliquots (500 µL)			
<input type="checkbox"/> Store at -80°C (until ship to C-BIG)		Date: ____/____/____	
<b>QUANTITY - ALIQUOTS</b>			
____ X vials at 500 µL			
<b>REAGENTS TRACKING</b>			
	Lot #	Expiry Date	Opened
Vacutainers	____	____/____	N/App
Finished N/App			
Notes:			

## Appendix 8.6: Sample Processing Form - RNA

Found in [Sample Processing Form – RNA (7Mar2022) CAPTURE ALS]




RNA Processing  
Sample Processing Form: RNA  
Last updated: 7March2022

RNA PROCESSING				SOP employed: CAPT-01-012	
Processed by: _____				CAPT ID: _____	
Time collected: _____				Date: ____/____/____	
VACCUTAINERS/TUBES			QUANTITY		
RECEIVED	TYPE		RECEIVED		
_____	_____		_____mL		
Issues	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/> Expired tubes Date of expiry: ____/____/____		
<b>STEPS:</b>					
<input type="checkbox"/>	1. Keep tubes upright at room temperature for a minimum of 2hours and a maximum of 72hours to allow blood to clot.				
<input type="checkbox"/>	2. Transfer tubes to -20°C for at least 24hours.				
<input type="checkbox"/>	3. Transfer tubes to -80°C freezer (until ship to C-BIG)			Date: ____/____/____	
REAGENTS TRACKING					
		Expiry Date	Opened	Finished	
Vacutainers	_____	____/____/____	N/App	N/App	

Notes:

## Appendix 8.7: Sample Processing Form – CSF

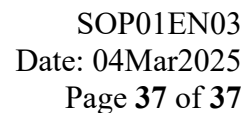
Found in [Sample Processing Form - CSF (7Mar2022) CAPTURE ALS]

		CSF Processing Sample Processing Form: CSF Last updated: 7March2022	
<b>CSF PROCESSING (SUPERNATANT)</b>			<b>SOP Employed: CBIG-02-008</b>
Processed by: _____		CAPT ID: _____	
Time start processing: ____:____		Date: ____/____/____	
<b>VACCUTAINERS/TUBES</b>		<b>QUANTITY</b>	
<b>RECEIVED</b>	<b>TYPE</b>	<b>RECEIVED</b>	
_____	_____	_____ mL	
<b>Issues</b>	<input type="checkbox"/> Blood contamination	<input type="checkbox"/> Not kept at 4°C immediately after lumbar puncture	<input type="checkbox"/> Expired tubes Date of expiry: ____/____/____
<b>STEPS:</b>			
<input type="checkbox"/> 1. Transfer the CSF to a new 15 mL Falcon tube			
<input type="checkbox"/> 2. Centrifuge for 10 minutes at 2000Xg		Start: ____:____	End: ____:____
<input type="checkbox"/> 3. Remove supernatant, and put it in a new 15 mL Falcon tube			
<input type="checkbox"/> 4. Remove supernatant and aliquot it.			
<input type="checkbox"/> 5. Prepare aliquots (500 µL)		Date: ____/____/____	
<input type="checkbox"/> 6. Store at -80 C (until ship to C-BIG)		Date: ____/____/____	
<b>QUANTITY - ALIQUOTS</b>			
_____ x 500µL			
Notes:			

## Appendix 8.8: Biosample Labels Template

Found in [Biosample Labels Template (EN, V2, 26Oct2022) CAPTURE ALS]

	Vial #	Type	VisitLabel	CollectionDate	Barcode	NewCode
CAPTXXXXXXXX	201	Serum	00M	DD/MM/20XX	CAPTXXXXXXXX201	XXXXXXXXYY
CAPTXXXXXXXX	202	Serum	00M	DD/MM/20XX	CAPTXXXXXXXX202	XXXXXXXXYY
CAPTXXXXXXXX	203	PBMC	00M	DD/MM/20XX	CAPTXXXXXXXX203	XXXXXXXXYY
CAPTXXXXXXXX	204	PBMC	00M	DD/MM/20XX	CAPTXXXXXXXX204	XXXXXXXXYY
CAPTXXXXXXXX	205	PBMC	00M	DD/MM/20XX	CAPTXXXXXXXX205	XXXXXXXXYY
CAPTXXXXXXXX	206	PBMC	00M	DD/MM/20XX	CAPTXXXXXXXX206	XXXXXXXXYY
CAPTXXXXXXXX	207	PBMC	00M	DD/MM/20XX	CAPTXXXXXXXX207	XXXXXXXXYY
CAPTXXXXXXXX	208	DNA	00M	DD/MM/20XX	CAPTXXXXXXXX208	XXXXXXXXYY
CAPTXXXXXXXX	209	DNA	00M	DD/MM/20XX	CAPTXXXXXXXX209	XXXXXXXXYY
CAPTXXXXXXXX	210	PLASMA	00M	DD/MM/20XX	CAPTXXXXXXXX210	XXXXXXXXYY
CAPTXXXXXXXX	211	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX211	XXXXXXXXYY
CAPTXXXXXXXX	212	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX212	XXXXXXXXYY
CAPTXXXXXXXX	213	RNA	00M	DD/MM/20XX	CAPTXXXXXXXX213	XXXXXXXXYY
CAPTXXXXXXXX	214	RNA	00M	DD/MM/20XX	CAPTXXXXXXXX214	XXXXXXXXYY
CAPTXXXXXXXX	215	Serum	00M	DD/MM/20XX	CAPTXXXXXXXX215	XXXXXXXXYY
CAPTXXXXXXXX	216	Serum	00M	DD/MM/20XX	CAPTXXXXXXXX216	XXXXXXXXYY
CAPTXXXXXXXX	217	Serum	00M	DD/MM/20XX	CAPTXXXXXXXX217	XXXXXXXXYY
CAPTXXXXXXXX	218	Serum	00M	DD/MM/20XX	CAPTXXXXXXXX218	XXXXXXXXYY
CAPTXXXXXXXX	219	Serum	00M	DD/MM/20XX	CAPTXXXXXXXX219	XXXXXXXXYY
CAPTXXXXXXXX	220	Serum	00M	DD/MM/20XX	CAPTXXXXXXXX220	XXXXXXXXYY
CAPTXXXXXXXX	221	Serum	00M	DD/MM/20XX	CAPTXXXXXXXX221	XXXXXXXXYY
CAPTXXXXXXXX	222	Serum	00M	DD/MM/20XX	CAPTXXXXXXXX222	XXXXXXXXYY
CAPTXXXXXXXX	223	Serum	00M	DD/MM/20XX	CAPTXXXXXXXX223	XXXXXXXXYY
CAPTXXXXXXXX	224	Serum	00M	DD/MM/20XX	CAPTXXXXXXXX224	XXXXXXXXYY
CAPTXXXXXXXX	225	Serum	00M	DD/MM/20XX	CAPTXXXXXXXX225	XXXXXXXXYY
CAPTXXXXXXXX	226	Serum	00M	DD/MM/20XX	CAPTXXXXXXXX226	XXXXXXXXYY
CAPTXXXXXXXX	227	Serum	00M	DD/MM/20XX	CAPTXXXXXXXX227	XXXXXXXXYY
CAPTXXXXXXXX	228	Serum	00M	DD/MM/20XX	CAPTXXXXXXXX228	XXXXXXXXYY
CAPTXXXXXXXX	229	Serum	00M	DD/MM/20XX	CAPTXXXXXXXX229	XXXXXXXXYY
CAPTXXXXXXXX	230	Plasma	00M	DD/MM/20XX	CAPTXXXXXXXX230	XXXXXXXXYY
CAPTXXXXXXXX	231	Plasma	00M	DD/MM/20XX	CAPTXXXXXXXX231	XXXXXXXXYY
CAPTXXXXXXXX	232	Plasma	00M	DD/MM/20XX	CAPTXXXXXXXX232	XXXXXXXXYY
CAPTXXXXXXXX	233	Plasma	00M	DD/MM/20XX	CAPTXXXXXXXX233	XXXXXXXXYY
CAPTXXXXXXXX	234	Plasma	00M	DD/MM/20XX	CAPTXXXXXXXX234	XXXXXXXXYY
CAPTXXXXXXXX	235	Plasma	00M	DD/MM/20XX	CAPTXXXXXXXX235	XXXXXXXXYY
CAPTXXXXXXXX	236	Plasma	00M	DD/MM/20XX	CAPTXXXXXXXX236	XXXXXXXXYY
CAPTXXXXXXXX	237	Plasma	00M	DD/MM/20XX	CAPTXXXXXXXX237	XXXXXXXXYY
CAPTXXXXXXXX	238	Plasma	00M	DD/MM/20XX	CAPTXXXXXXXX238	XXXXXXXXYY
CAPTXXXXXXXX	239	Plasma	00M	DD/MM/20XX	CAPTXXXXXXXX239	XXXXXXXXYY
CAPTXXXXXXXX	240	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX240	XXXXXXXXYY
CAPTXXXXXXXX	241	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX241	XXXXXXXXYY
CAPTXXXXXXXX	242	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX242	XXXXXXXXYY
CAPTXXXXXXXX	243	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX243	XXXXXXXXYY
CAPTXXXXXXXX	244	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX244	XXXXXXXXYY
CAPTXXXXXXXX	245	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX245	XXXXXXXXYY
CAPTXXXXXXXX	246	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX246	XXXXXXXXYY
CAPTXXXXXXXX	247	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX247	XXXXXXXXYY
CAPTXXXXXXXX	248	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX248	XXXXXXXXYY
CAPTXXXXXXXX	249	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX249	XXXXXXXXYY
CAPTXXXXXXXX	250	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX250	XXXXXXXXYY
CAPTXXXXXXXX	251	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX251	XXXXXXXXYY
CAPTXXXXXXXX	252	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX252	XXXXXXXXYY
CAPTXXXXXXXX	253	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX253	XXXXXXXXYY
CAPTXXXXXXXX	254	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX254	XXXXXXXXYY
CAPTXXXXXXXX	255	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX255	XXXXXXXXYY
CAPTXXXXXXXX	256	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX256	XXXXXXXXYY
CAPTXXXXXXXX	257	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX257	XXXXXXXXYY
CAPTXXXXXXXX	258	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX258	XXXXXXXXYY
CAPTXXXXXXXX	259	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX259	XXXXXXXXYY
CAPTXXXXXXXX	260	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX260	XXXXXXXXYY
CAPTXXXXXXXX	261	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX261	XXXXXXXXYY
CAPTXXXXXXXX	262	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX262	XXXXXXXXYY
CAPTXXXXXXXX	263	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX263	XXXXXXXXYY
CAPTXXXXXXXX	264	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX264	XXXXXXXXYY
CAPTXXXXXXXX	265	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX265	XXXXXXXXYY
CAPTXXXXXXXX	266	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX266	XXXXXXXXYY
CAPTXXXXXXXX	267	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX267	XXXXXXXXYY
CAPTXXXXXXXX	268	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX268	XXXXXXXXYY
CAPTXXXXXXXX	269	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX269	XXXXXXXXYY
CAPTXXXXXXXX	270	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX270	XXXXXXXXYY
CAPTXXXXXXXX	271	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX271	XXXXXXXXYY
CAPTXXXXXXXX	272	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX272	XXXXXXXXYY
CAPTXXXXXXXX	273	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX273	XXXXXXXXYY
CAPTXXXXXXXX	274	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX274	XXXXXXXXYY
CAPTXXXXXXXX	275	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX275	XXXXXXXXYY
CAPTXXXXXXXX	276	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX276	XXXXXXXXYY
CAPTXXXXXXXX	277	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX277	XXXXXXXXYY
CAPTXXXXXXXX	278	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX278	XXXXXXXXYY
CAPTXXXXXXXX	279	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX279	XXXXXXXXYY
CAPTXXXXXXXX	280	CSF	00M	DD/MM/20XX	CAPTXXXXXXXX280	XXXXXXXXYY



Found in [Sample Packing Slip (12July2022) CAPTURE ALS]

## CAPTURE ALS