National Institute of Environmental Health Sciences

SOCIAL & SCIENTIFIC SYSTEMS, INC.
an employee-owned company

Clinic Site Specimen Processing Procedure

Document No.:	Supersedes Document No.:	Issue Date:	Effective Date:
Process Owner Approval:	**Title:**	**Signature:**	**Date:**
Functional Area Approval:	**Title:**	**Signature:**	**Date:**

Track Changes	

. PURPOSE

The purpose of this standard operating procedure (SOP) is to describe how to process the blood samples collected at clinics within the system.

. SCOPE AND SAFETY PRECAUTIONS

This SOP applies to all clinic staff that performs tasks related to the processing of samples from the clinic. Exercise Universal Precautions.

. EQUIPMENT

Disposable gloves	Transfer Pipettes (disposable)
Lab Coats	2mL Cryovial racks
Tourniquet (latex-free) BD	8mL Vial Racks
Alcohol wipes BD	Freezer Storage Boxes (2” and 3”)
2”x2”gauze pads	Grids (9X9)
1 inch surgical tape	Grids (7X7)
21g butterfly needle blood collection with 12” tubing and adapter	Supplies for handling dry ice (e.g. scoop, insulated tray, heavy gloves)
Band-Aids	Dry Ice
10 ml Red Top Vacutainer	Absorbent sheets for bench top
10 ml Lavender Top EDTA Vacutainer	Biohazard bags and boxes
Tube rack (holds 72 10-13mm test tubes	Specimen Transport Bags
Sharps container 1 Gal	Fed-Ex air bills, air transport labels

2.0 mL Thermo Cryovials #430526	Packing tape strips
Sarstedt 8mL Tubes #60.542.007	Hand sanitizer (if necessary)
Transfer Pipettes (disposable)	

. BLOOD PROCESSING

Label Aliquots

Label all aliquot tubes with the labels, shipped by SSS to the protocol site, before aliquotting.

Note: The labels do not have the Subject Id on them. The BSI Id on the label is linked to the subject Id in the protocol management system (SMS).

Label sets of five 2mL Thermo cryovials for each red top and seven 2mL Thermo cryovials for the lavender top from a given protocol participant using the Serum and Plasma/RBC labels from each participant's label set. Place the labeled cryovials in the plastic rack as shown in the diagram below.

[Note: the following sections are included if the clinic sites process samples, as specified in the protocol.]

Plasma 0401	**Plasma 0402**	**Plasma 0403**	**Plasma 0404**	**Plasma 0405**	**RBC/Buffy 0411**	**RBC/Buffy 0412**
Serum 0301	**Serum 0302**	**Serum 0303**	**Serum 0304**	**Serum 0305**		
Serum 0201	**Serum 0202**	**Serum 0203**	**Serum 0204**	**Serum 0205**		
Serum 0101	**Serum 0102**	**Serum 0103**	**Serum 0104**	**Serum 0105**		
Empty	Empty	Empty	Empty	Empty		

Label one 8 mL Sarstedt tube per red top tube collected from each protocol participant using the Clot labels from each participant's Label Set. Place the labeled 8 mL tubes in a blue wire rack as shown in the diagram below.

Clot 0110	**Clot 0210**	**Clot 0310**
Empty	Empty	Empty

Centrifuge the Red Top and Lavender Tops

Always use Universal Precautions. Process blood specimens within 6 hours of collection if at all possible.

Place Lavender Top Vacutainers on a rocker for 5 minutes or mix well by inversion before centrifugation.

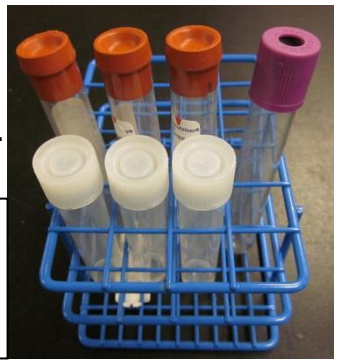
Place the red top and the lavender top tubes in the centrifuge so that they are cross-balanced in the cups. If there are an odd number of tubes to spin, counterbalance by filling the empty position(s) with a BALANCE tube filled with water.

Set up the centrifuge to spin at the following settings:

- RCF (Avg) = 800
- RCF (Max) = 1200
- Time = 00:15
- Acceleration = MAX
- Deceleration = MAX
- Temp = 24°C

.8.1 **Spin the Red and Lavender Top Tubes** for 15 min at 1200 RCF. Note start *and stop times in the SMS.*

.8.2 **Remove the Red Top Tubes** from the centrifuge and place them in the blue wire rack as shown in the photo below. Proceed to Step 4.4.



Figure/Diagram 1: placement of tubes in racks for processing

Processing Flow Chart

Collect		Processing	Sample Type	# Aliquots	Size of Aliquot	Vial Size & Type	Box Config	# / Box	Store Temp
1	**10 ml Red Top #366430** ***Collection 1***	Centrifuge at +4C	Serum	5	1mL	2ml Thermo Cryovial	5"x5"x3" Box - 9x9 grid	81	-80°C
			Clot	1	5mL	8ml Sarstedt Cryotube	5"x5"x3" Box - 7x7 grid staggered	42	-80°C
2	**10 ml Red Top #366430** ***Collection 2***	Centrifuge at +4C	Serum	5	1mL	2ml Thermo Cryovial	5"x5"x3" Box - 9x9 grid	81	-80°C
			Clot	1	5mL	8ml Sarstedt Cryotube	5"x5"x3" Box - 7x7 grid staggered	42	-80°C
2	**10 ml Red Top #366430** ***Collection 2***	Centrifuge at +4C	Serum	5	1mL	2ml Thermo Cryovial	5"x5"x3" Box - 9x9 grid	81	-80°C
			Clot	1	5mL	8ml Sarstedt Cryotube	5"x5"x3" Box - 7x7 grid staggered	42	-80°C
4	**10 ml Lavender Top #366643** ***Lav. Collection***	Centrifuge at +4C	Plasma	4-5	1.5mL	2ml Thermo Cryovial	5"x5"x3" Box - 9x9 grid	81	-80C
			RBC/Buffy	2	1.5mL	2ml Thermo Cryovial	5"x5"x3" Box - 9x9 grid	81	-80C

RED TOP TUBES: SERUM AND CLOT PROCESSING

After centrifuging the red tops, uncap the cryovials and Sarstedt tubes for a single vacutainer and place the caps top down on a clean KimWipe.

Dispense 1.0 mL serum, using a new transfer pipette, for each participant from red top tube ***RED Collection 1*** into the corresponding set of five cryovials for that participant. Move each row forward as you fill them.

NOTE: because of incomplete specimen collection or sample recovery, you may not be able to dispense a full 1.0 mL of serum into all aliquot tubes. Fill as many tubes as possible. Record low volume if the final aliquot is less than 0.5 mL.

Pour over the clot into the corresponding labeled Sarstedt tubes (***CLT 0110***).

Tightly recap all of the 1 mL serum cryovials and the 8 mL clot tube.

Repeat 4.4.2 & 4.4.3 for all remaining participants' red top tubes.

Note in the data management application any low-volume aliquots and the rack of tubes into the refrigerator until the end of the day or when you can box the samples.

Remove the labels from the empty serum cryovials and place them on a discard sheet. **Discard** the empty red top tubes in the biohazardous waste container.

After completing all processing for a participant, following the Boxing Procedures in Section 5 to transfer all of the serum and clot aliquots into the appropriate cryostorage boxes for freezing.

Freeze all samples at -80°C

LAVENDER TOP TUBES: PLASMA AND RBC/BUFFY PROCESSING

Uncap the cryovials (**Plasma 0401-0405** and **RBC/Buffy 0410-0411**) and for the Lavender Top **Lav. Collection 0310** vacutainer and place the caps top down on a clean KimWipe.

Dispense 1.0 mL Plasma, using a new transfer pipette, for each participant from Lavender Top **Lav. Collection 0310** vacutainer into the corresponding set of five cryovials (**Plasma 0401-0405**) being careful not to disturb the RBC/Buffy layer for that participant. Move each row forward as you fill them. After you have dispensed the plasma, dispense 1mL of the RBC/Buffy layer in two cryovials (**RBS/Buffy 0410-0411**). You will most likely have RBC/Buffy layer remainder.

NOTE: Low Volumes: See note above in serum aliquotting section.

Tightly recap all of the cryovials.

Note in the data management application any low-volume aliquots and place the rack of tubes into the refrigerator until the end of the day or when you can box the samples.

Remove the labels from the empty lavender top cryovials and place them on a discard sheet. **Discard** the empty lavender top cryovials in the biohazardous waste container.

After completing all processing for a participant, following the Boxing Procedures in Section 5 to transfer all of the Plasma and RBC/Buffy aliquots into the appropriate cryostorage boxes for freezing.

Freeze all samples at -80°C.

Processing Cracked Vials

For cracked or leaking red top and lavender top vacutainers, you must be concerned about the integrity of the sample. If the Vacutainer has cracked you must be concerned with leaking and if any pieces of glass may be in the sample. If the tube is cracked or leaking pour over the entire Vacutainer into a 15mL centrifuge tube and centrifuge.

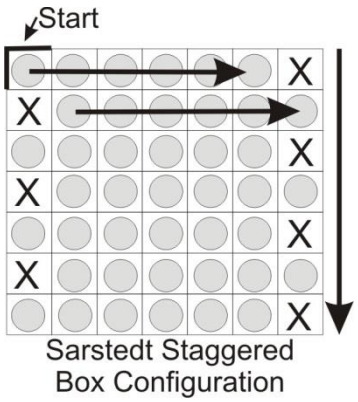
. BOXING PROCEDURES

2ml Thermo Cryovial #430526	5”x5”x2” Box - 9x9 grid
8ml Sarstedt Cryotube #60.542.007	5”x5”x3” Box - 7x7 grid staggered

Label boxes with both a box number and a material type using the labels provided.

2ml Thermo Cryovials will be stored in 5”x5”x2” Boxes with a 9x9 grid. Mark the back left corner of the box with a sharpie and fill all spots in the box.

8mL Sarstedt vials will be stored in 5”x5”x3” Boxes with a 7x7 grid. They will be placed in a staggered configuration leaving one space open in each row (See diagram).



Rubber band each box before placing it in the freezer.

Store the boxes in a -80°C freezer. Freeze samples for at least 12 hours prior to shipment.