Alberta Health Services
Laboratory Services

Special Coagulation Aliquoting

This document is applicable at site(s):
All Sites

Applicability This document applies to all personnel of AHS Laboratory Services, the Lamont Health Centre and laboratories administered by Covenant Health.

Purpose This document provides instruction on special coagulation aliquoting.

Procedure **A. Aliquoting Chart**
NOTE: A requisition must accompany the sample for all test codes indicated with an asterisks (*)

Test Code	Test Description	Acceptable Specimen Tube Reject all other specimen types	Frozen Aliquots Plasma = citrated
***APA**	Antiphospholipid Antibodies or Cardiolipin Antibodies	• Gold top tube • Ordered in conjunction with LUP.	2 x 0.5 ml **serum** Mark **serum** on label
***APCE**	APC Resistance or Activated Protein C Resistance or clot based Factor V leiden	• Glass/Plastic blue top tube	2 x 1.0 ml plasma
APCGN and/or PROM	Factor V Leiden mutation and/or Prothrombin 20210 (or Factor II) mutation	• 1 – 4 mL mauve top tube @ 4ºC • MUST be sterile, dedicated sample	N/A – MUST be submitted as whole blood. Send Cold.
COAGZ	Special Coagulation Evaluation: This is a Morphology/Coagulation Pathologist On-Call initiated order - they will contact UAH Special Coag with specific test order	• Glass/Plastic blue top tube	3 x 0.5 ml **Platelet Free Plasma** min. Mark **PFP** on label
***F2**	Factor II	• Glass/Plastic blue top tube	2 x 0.5 ml plasma per test
***F5**	Factor V		
***F7**	Factor VII		
***F8**	Factor VIII		
***F8INH**	Factor VIII Inhibitor	• Glass/Plastic blue top tube	2 x 1.0 ml plasma
***RISTO**	Ristocetin Cofactor	• Glass/Plastic blue top tube	2 x 0.5 ml plasma per test
***VWFAG**	Von Willebrand Factor **or** VWF Antigen		
***F9**	Factor IX	• Glass/Plastic blue top tube	2 x 0.5 ml plasma per test
***F10**	Factor X		
***F11**	Factor XI		
***F12**	Factor XII		
***F13**	Factor XIII	• Glass/Plastic blue top tube	2 x 1.0 ml plasma
***HITB**	Heparin induced thrombocytopenia assay	• Gold top tube	2 x 0.5 ml **serum** Mark **serum** on label
***LUP**	Lupus anticoagulant **or** Dilute Russell's Viper Venom Time	• 2 GLASS blue top tubes • **Plastic WILL NOT be accepted** • Ordered in conjunction with APA	2 x 1.0 ml **Platelet Free Plasma** Mark **PFP** on label
HEPRN	Low Molecular Weight Heparin **or** Anti-Xa assay **or** Unfractionated Heparin (LMWH: Enoxaparin, Fragmin, Dalteparin)	• Glass/Plastic blue top tube **– must be collected** 4-6 hours post dose. **Room Temp**.	MUST be processed within 4 hrs of collection. 2 x 0.5 ml **Platelet Free Plasma** Mark **PFP** on label
AT3	Antithrombin	• 1 Glass/Plastic blue top tube	1 x 0.5 mL plasma
***PROTC**	Protein C	• 2 Glass/Plastic blue top tubes	2 x 0.5 ml plasma per test
***PROTS**	Protein S		
***PTIN**	PT Inhibitor Screen	• 2 Glass/Plastic blue top tubes	2 x 2.0 ml plasma
***PTTIN**	PTT Inhibitor Screen	• 3 Glass/Plastic blue top tubes	2 x 3.0 ml **Platelet Free Plasma** Mark **PFP** on label
TT	Thrombin Time	• 1 Glass/Plastic blue top tube	2 x 0.5 ml plasma

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B. Aliquoting Instructions

Step	Detail	Information
1. Spin specimen.	1.1) 2500 g/rcf for 10 minutes. 1.2) If specimen received spun proceed to **Step 2**. 1.3) **Aliquot and freeze sample aliquots within 4 hrs of collection.**	• Platelet poor plasma must have a platelet count of $<10X10^9$/L.
2. Check specimen integrity before pooling.	2.1) Ensure patient identification for all collection tubes are the same. 2.2) Using a **plastic** pipette, remove the plasma or serum from cells one tube at a time – avoid the platelet and WBC layer when removing plasma and/or clot separator when removing serum.	**NOTE:** Check for a clot by rimming the red cell button with an applicator stick.

If…	Then…
Removing plasma	• Keep plasma in pipette. • Check sample for a clot prior to adding to the pool.

If…	Then…
Clotted	• DO NOT add to the plasma pool. • Discard plasma.
Hemolyzed	• Freeze hemolyzed plasma separately and label as HEMO.
Not clotted and not hemolyzed	• Proceed to **Step 3**.

If…	Then…
Removing serum	
Hemolyzed	• Freeze hemolyzed plasma separately and label as HEMO.
Not hemolyzed	• Proceed to **Step 3.**

3. Pool plasma and/or serum.	3.1) Into plastic container.	• Accuvette or sterile urine container.
4. Transfer the pooled plasma.	4.1) Mix the pool, aliquot into labeled plastic tubes, and freeze at -70ºC – refer to **Section A**. ** **Preferred tube:** 4 mL VWR #718585 and screw top lid VWR #718593 **or** refer to *Client Supply Order – Edmonton Zone Form***	**NOTE:** Sample may be stored at -20ºC for a short period (eg. 2 days) in a NON-frost free freezer; sample should be shipped on dry ice.

C. Preparation of Platelet Free Plasma (PFP)

Step	Detail	Information
1. Perform the primary spin.	1.1) Follow steps 1-3 in **Section B**.	
2. Perform a secondary spin.	2.1) Mix the pool and aliquot into labeled plastic eppendorf tubes. 2.2) Spin in a high speed centrifuge for 6 mins. **NOTE: If a high speed centrifuge is not available, transfer plasma to plastic tubes and repeat Step 1 in Section B.**	• UAH (Eppendorf 5415D – 12000 rpm /13600 g/rcf – 6 min) • RAH (Eppendorf 5452 – 13400 rpm – 6 min)
3. Label aliquot tubes.	3.1) Indicate on the label that it is **PFP** on appropriate number of tubes. **NOTE:** Aliquot tubes must be plastic – refer to **Section B, Step 4.**	
4. Aliquot PFP.	4.1) Remove plasma with **plastic** pipette and pool – be careful not to disturb the cell button. 4.2) Mix the pool, aliquot into labeled plastic tubes, and freeze at -70ºC – refer to **Section A**.	**NOTE:** Sample may be stored at -20ºC for a short period (ie. 2 days) in a NON-frost free freezer; sample should be shipped on dry ice.

References N/A

Related Documents Current Version of:

Document	Document Control Number
Client Supply Order – Edmonton Zone Form	RRRSOR00005

Contact Information University of Alberta Hospital Routine Coagulation (780) 407-7232