OSUCCC LTB Laboratories Procedure Preparation of plasma and serum from peripheral blood[1]			Effective: 10/01/1997
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Procedure No.	Supercedes:	Created/Review Date:	Number of Pages:
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1.0 PRINCIPLE

Specimens of plasma and serum can be examined to ascertain the concentrations of proteins, chemicals or treatment drugs in the blood, especially in correlation with a time point in a treatment regimen. The specimens can be stored for later data collection and analysis if samples are prepared and frozen shortly after acquisition.

Plasma is the liquid portion of blood complete with all proteins. Plasma may be prepared from any blood collection tube containing an anti-coagulant including: Sodium heparin, lithium heparin, EDTA, &c.[1]

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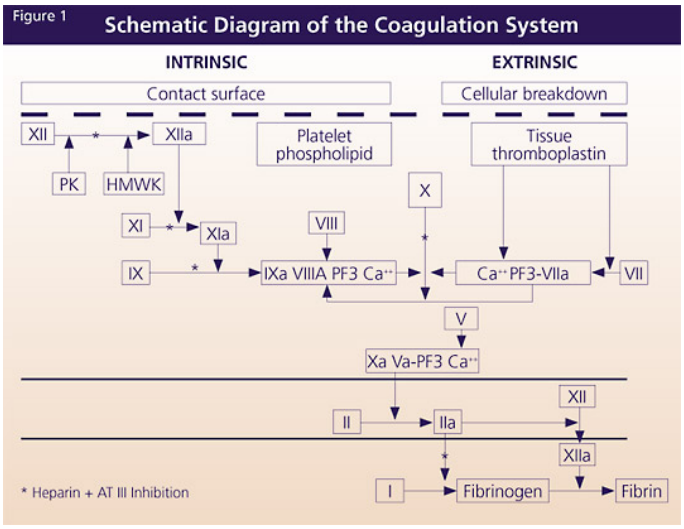

Serum is prepared from blood that has been allowed to clot and therefore contains no fibrinogen. Serum is prepared from blood collection tubes with no additive, sometimes with a serum separator gel plug.

For samples prepared prior to receipt in the LTB, please see the relevant protocol for previously prepared samples.

2.0 SPECIMEN

Specimens should be procured as soon as they are received by the lab and preferably within 24 hours of being procured from the patient/subject. Patient samples that require plasma preparation and viable cell procurement must be held at room temperature to insure viability of cells. If samples are rec'd on ice or icepacks, plasma may be prepared but cell viability will be damaged and cells should not be procured. When samples are incorrectly prepared, notify the study contact listed in the on sample collection forms as soon as possible. Samples for all ALLIANCE studies should be collected into the appropriate collection tube indicated in the study protocol. Any problems or comments regarding sample collection, shipment and receipt will be noted by the technician on the sample procurement form. For non-ALLIANCE study samples, the Procurement lab will be notified that a patient sample is waiting in the 2nd floor Oncology lab, James Cancer Hospital for processing.

2.1 For plasma isolation: Fresh peripheral blood collected with anticoagulation reagent (heparin, EDTA) that is specified in the sample collection protocol.

2.2 For serum isolation: Fresh peripheral blood collected with no anticoagulation agent. Collection tube may contain gel plug.

3.0 MATERIALS AND SUPPLIES

Sterile pipets (5ml, 10ml and 25ml)
Sterile 4x4 gauze
70% isopropyl alcohol
Sterile cryovials (2ml)
Pipet-aid

4.0 EQUIPMENT

Biosafety cabinet
Benchtop centrifuge with swinging bucket rotors to hold 6 and 10cc vacutainer blood collection tubes. Buckets should be equipped with dome covers to contain potentially biohazardous specimens.

5.0 QUALITY CONTROL AND SAFETY

It is recommended that specimen collection be carried out in accordance with NCCLS document M29T2. No known test sample can offer complete assurance that human blood samples will not transmit infection. Therefore, all derivatives are potentially infectious. Always spray alcohol on the caps before opening solutions. The alcohol can be dried off using gauze. If you have been out of the hood for a while and are wearing the same pair of gloves, use a new pair of gloves. Remember not to touch the sides (inside or outside) of any bottles with your pipette – if you do, dispose of the pipette and start again.

6.0 PROCEDURE

6.0.1 Samples are received and recorded onto the study worksheet.

6.0.2 Prepare plasma or serum by centrifugation of a **peripheral blood** sample at 2500 rpm for 10 minutes (**Plasma and Serum are *never* procured from bone marrow samples.**)

6.0.3 Transfer 1 ml of plasma into each cryovial, preparing the appropriate number of plasma/serum vials as required by the treatment or correlative collection protocol. Red caps are used for serum and Purple caps are used for plasma. Cryovials are labeled with patient accession ID only.

6.0.4 Plasma and serum are stored at –80°C until requested for analysis.

6.0.5 If applicable, process residual sample for viable cells using standard procurement procedure.

7.0 LIMITATIONS OF THE PROCEDURE

Centrifugation is the process of separating lighter portions of a mixture or suspension from heavier portions by centrifugal force based on their relative densities. The separation of anticoagulated blood components by centrifugation is driven by differences in density and cell size. The heavier and larger red blood cells and white blood cells sediment more quickly than platelets. Hence, platelets are the primary cell type that can be found in plasma and the plasma obtained under most recommended centrifugation conditions used in chemistry is not completely acellular. The centrifuge speed, time and temperature, as well as patient cell counts, can influence the purity of the plasma or serum.[2]

8.0 REFERENCES

1. NCCLS, Evacuated Tubes and Additive for Blood Specimen Collection – Fourth Edition; Approved Standard. NCCLS documentation H1-A4, Vol. 16, No. 13 Dec. 1996.

2. Becton Dickinson LabNotes - Volume 13, No.2, Spring 2003.