

Section 10.22

Procedure for separating Plasma and Serum from whole blood

Aim: Effective Separation of blood products

Purpose: To standardize separating procedures so that research samples will be uniform in quality

The decision to collect anticoagulated (plasma, buffy coat, RBC) or coagulated (serum, clot) blood samples must be made prior to collection so that proper blood draw tubes will be used.

Serum (needs clot time)

A serum separator tube (SST, tiger top tube). Let the blood sit for 30 minutes to one hour at room temperature to clot before spinning and separating.

A delay in centrifugation may have a detrimental effect on the sample quality and may result in inaccurate results. Avoid hemolysis.

Separating plasma (time sensitive)

Tube with an anti coagulant eg: Edta (lavender top) sodium heparin (green top), sodium citrate (blue top) are used for separating Plasma You need to spin and separate within one hour of receiving the specimen (time sensitive)

Note:

Universal Precautions must be used when working with blood. Use of personnel protective equipment is mandatory. Use of eye protection is mandatory unless blood tubes are being opened and serum/plasma/whole blood are being aliquoted inside a BL2 safety cabinet.

***Keep blood on wet ice and process within one hour of blood draw**

Separation of plasma

- 1) Blood will be collected into purple top EDTA tubes and centrifuged (2000 rpm) at 4 degrees centigrade for 20 minutes.
- 2) After centrifugation using clean pipette technique place 1.0ml of plasma into 1.5ml eppendorf tube labeled with tracking number and "plasma"
- 3) Freeze immediately at -80 degree freezer

Separation of Serum

1. A 10 ml tube of whole blood will be collected following standard procedures using a serum separator tube (SST, tiger top tube) from each patient.
2. Allow samples to clot for one hour at room temperature
3. Centrifuge for 10 minutes at approximately 1000g
4. Using clean pipette technique Aliquot 210ul of serum into labeled cryovials.
5. Immediately freeze vials of serum at -80-degree freezer

Aliquoting whole blood

Whole blood will be aliquoted into sterile tubes upon receipt by carefully inverting the blood tube so that it is gently mixed before pipetting appropriate amounts (protocol specific) of whole blood into appropriate storage tubes using clean pipette tips between each patient.

- Gently invert the tube of blood to mix contents
- Carefully open blood tube (universal precautions; gloves, eye protection)
- With clean pipette tip aliquot appropriate amount of whole blood into clean/labeled storage tubes.
- Freeze in -80 freezer