



**Intended Use**

To delineate procedures for processing of blood, urine, CSF, stool and body fluid specimens to minimize preanalytical variability and ensure accurate test results.

**Specimen Containers**

1. The chemistry department processes many different types of specimen collection tubes.

Specimen Container	Additive
SST yellow top 13 X 75 and 13 X 100 (serum)	Clot activator and gel for serum separation. Silicone coated interior
PST Light Green Top 13 X 75 (plasma)	Lithium heparin (spray-coated) and polymer separator gel
Gray top 13 X 75 (plasma)	Sodium fluoride & Potassium oxalate
Red top 13 X 75 (serum)	Clot activator and silicone coated interior
Clear Cap 13 X 75	No additives
Lavender top 13 X 75 (plasma)	K2 EDTA (spray-dried)
Microtainer SST with yellow cap	Serum separator
Microtainer PST with light green cap	Lithium heparin and plasma separator gel
Terumo Capiject Gray Top Micro Container	NaFl & Potassium oxalate
Microtainer Tube with red cap	None
Microtainer Tube with purple cap	Dipotassium EDTA
Yellow top 16mm X 100 mm X 8 mL BD Vacutainer® plus plastic urinalysis tube with conical bottom	Sterile evacuated tube with no additives
Cherry red / Yellow top 16mm X 100 mm X 8 mL BD Vacutainer® plus conical plastic urinalysis preservative tube	Sterile evacuated tube with preservative (Unacceptable for any testing other than urinalysis)

2. Cerebral spinal fluid (CSF) is usually collected in LP sterile collection tubes. Tube #1 is the preferred tube for chemistry testing.
3. Urine samples for Urinalysis may be collected in either of the conical tubes listed above, or in sterile screw cap containers (120 mL blue-top or white top). Urine from newborns may be received in pedibags.
4. 24 hour urine chemistry collections are usually received in 3000 ml plastic urine collection jugs and must be refrigerated during collection.
5. Body fluids are usually received in various types of containers and tubes and may need to be transferred to a clear cap BD 13 X 75 specimen tube that can be sampled by the chemistry analyzer. This tube is recommended for all urine chemistries, fluids and DAUs.
6. Study samples - The Chemistry department is notified (Supervisor and/or Specialist) prior to arrival and instructions are given as to use of containers, sample type and labeling.

## Equipment

### 1. Centrifuges

Refrigerated and non-refrigerated – sample tubes with the same volumes are symmetrically loaded

- a. Horizontal-head or swinging-bucket
- b. Angle-head rotor or fixed angle
- c. Microfuge – fixed angle for microtainers and bullets
- d. Ultracentrifuge – very high speed fixed-head rotor for clearing serum of chylomicrons

### 2. Materials

- a. Gloves
- b. Laboratory coats or aprons
- c. Laboratory disinfectant (for counters, centrifuges and spills)
- d. Sharps disposal containers
- e. Biohazard disposal cans with red bag liners
- f. Processing shields or glasses
- g. Masks
- h. Gauze pads
- i. Cotton swabs
- j. Applicator sticks
- k. Plastic falcon tubes and caps
- l. Microtainer plastic insert extenders
- m. Short sample falcon tube inserts
- n. Parafilm
- o. Aluminum foil
- p. Pen with indelible water proof ink

## Quality Control

All centrifuges are periodically checked by the Quality Assurance department for RPM accuracy.

## Procedure

### Labeling

1. Specimen tubes are checked for correct labeling (minimum requirements: complete name of patient and unit number or date of birth) and collection information. Mislabeled or unlabeled specimens are returned to SARC with notation as to same in specimen bag. Alternately, these specimens may be given to the section supervisor.
2. EMR ordered samples should arrive bar-coded and received. The bar code label should have the collection time and initials of the collector on it. An exception is made for tubes demographically labeled, with collection information on the tube with the LIS label accompanying it. (microtainers, etc)
3. The bar-code label placed on a tube should have the bars follow one another down the tube (“ladder placement”). The label should not be skewed more than  $\pm 5\%$ . The bottom of the label should be 0.34 inches (0.86 cm) from the bottom of the tube. There should also be a minimum quiet zone (blank, white space on top and bottom of bar-code bars) of 0.20 inches (0.51 cm) at each end of the barcode symbol. (No writing or initialing in the quiet zone.) Torn, marked, wet and poorly printed barcodes should be reprinted. The recommendation applies to both primary and aliquot tubes used for direct sampling on analyzers.
4. Labels should be placed as high as possible on the tube without touching the cap and not covering the entire tube, so that the sample can be seen from the side of the tube.
5. Samples not requiring bar code labels should generally follow the above labeling instructions.

## Processing

### Serum and Plasma Specimens

1. Serum samples should be allowed to completely clot before centrifugation. Tubes should be placed in a vertical position and allowed to clot. Becton-Dickinson recommends the following minimum clot times for their tubes. **Serum samples must be completely clotted (red cell clot has retracted) before centrifugation.** Samples that were not allowed to clot vertically may need to be rimmed to remove fibrin and clots adhering to the stopper. Specimen tubes not listed below should be held a minimum of 30 minutes prior to centrifugation.

Collection Tube	Clotting Time*
PLUS(plastic) serum tubes - Red Tops	60 minutes
PLUS(plastic) tubes - SST	30 minutes
SST & Red cap microtainers	30 minutes

\*Clotting time is the total time from time of draw.

2. All anti-coagulated samples (PST, lavender top and gray top tubes and microtainers) should not be clotted. These samples should be checked for clots prior to centrifugation and any clotted samples should be canceled.
3. Specimens will be centrifuged according to the recommended times by Beckman Coulter as they would be centrifuged on the automation line and from a study performed at UCDCM. **All unspun tubes should be mixed 5-10 times before centrifugation.** Tubes received from clinics already centrifuged and not transported upright will be re-centrifuged for one minute. Fibrin clots found should be removed first before re-centrifuging. **Do not centrifuge previously centrifuged tubes with unspun tubes.**

Collection Tube	RCF (xg)	Centrifugation Time
PLUS(plastic) SST & PST***	2100	4 minutes
PLUS(plastic) SST & PST***	4400	3 minutes
Non-gel red top, gray top, purple top & Clear Cap tubes	2100	4 minutes
Non-gel red top, gray top, purple top & Clear Cap tubes	4400	3 minutes
All microtainers with extenders	2100	4 minutes
All microtainers with extenders	4400	3 minutes

\*\*\*The gel used in the SST tubes contains a polymer barrier material with a specific gravity between that of the serum/plasma and blood cells. During centrifugation of the blood specimen, the polymer barrier material rises to the interface of the serum/plasma and the clot/cells and forms a physical barrier separating the serum/plasma from the clot/cells.

RPM – Revolutions per minute – speed at which the centrifuge rotor spins

RCF – Relative centrifugal force – also referred to as g-force (xg)


The applied force resulting from the spinning action that is perpendicular to the axis of rotation.

g – Gravity – Universal constant that represents the natural pull or force of objects toward earth.

The speed at which the rotor spins produces a centrifugal force that follows the relationship,  $RCF = 0.0001118 \times \text{radius} \times \text{RPM}^2$ . This means that RCF increases exponentially with an increase in RPM. Therefore **RPM** and **RCF** are not the same.

4. All labels used for aliquoted samples must have the following noted on the label.
  - initials of processor
  - time of collection



- type of specimen (red, green, etc)
5. Aliquoting is done using a clean plastic transfer pipette. Re-verify name on original tube vs aliquot tube prior to transferring contents. Cap tubes securely. Minimum volume requirements, specimen requirements, and storage conditions are available in the [Specimen Processing Requirements Manual](#) on the processing bench.
  6. Grossly lipemic samples may need to be ultracentrifuged prior to analysis. (If testing includes a lipid panel, an aliquot is saved to dilute triglyceride, cholesterol and HDL.) Refer to the Beckman Airfuge Ultracentrifuge operating procedure for instructions on loading and the required amount for proper operation. Ultracentrifugation is limited to those with proper training to perform this task.
  -  7. After centrifugation, the degree of hemolysis should be within allowable limits for the testing requested. Any degree of hemolysis is unacceptable for some specimens (VitD, Fe, Folate, PEP, **Ammonia**, etc). Specimens are visually examined for hemolysis. The hemolysis chart can be used for comparison to aid in determining the degree of hemolysis. **Samples run on the automation line will have a hemolysis index performed. More information about serum indices can be found in the Sample Indices Technical Procedure 3035. Grossly hemolyzed Chemistry I samples are not acceptable and should be canceled. Grossly hemolyzed samples are only accepted for the first 24 hours for a post burn admission patient.**
  8. For the minimum volume requirements and recommended aliquot tubes when aliquoting samples for Chemistry, Special Chemistry and Immunology, refer to the specimen requirements sheet for sample processing in the Manual Processing Information binder at the Processing Bench.

### Urine samples

1. Spot urine chemistry samples should be received in a separate clear cap tube even if a urinalysis has been ordered. If the urine sample submitted is in a bottle, the sample is aliquoted into a 13 X 75 clear cap tube for analysis at the UA bench. Urine for Toxicology requires 3 clear cap tubes of urine
2. Timed urine samples will be submitted with a requisition request with documented start date and time, stop date and time, and height and weight if a creatinine clearance is ordered. If no requisition slip was submitted and all information is written on the containers, use the 24 hour urine worksheets that are next to the hood to transcribe collection information including total volume. Total urine volume (in mL) of each container can be read from the container and added together to obtain the total volume. When aliquoting, multiple containers must be mixed together before taking a representative sample aliquot. An extra aliquot (BD UA tube) should be saved in the Sendout refrigerator with the patient's specimen label and all collection information and total volume written on it.

### Procedure Notes

1. Specimens failing acceptable criteria for any reason (hemolysis, volume, type of anticoagulant) are rejected for analysis. The inpatient, ER and day shift outpatient STATs will have the clinic area responsible for specimen collection notified by phone as soon as possible. After-hours clinic notification is via rejection documentation in LIS. All canceled specimens are documented in the LIS as per LIS Specimen Cancellation Procedure. Unlabelled/mislabelled specimens: place specimen, order slip and canceled label in a specimen bag and put it in the chem walk-in refrigerator UN/MIS BOX. All other cancellations: write "canceled" across the bar code, place specimen in rear area of chemistry white rack.
2. Practice Universal precautions at all times.
3. Handle all biologic samples and equipment according to departmental policy.
4. All biohazardous specimens and equipment are disposed of in approved biohazard containers. Non-sharps are placed in red bag containers for disposal. Sharps are disposed of in "sharps" containers.
5. All paperwork that contains patient information is placed in the white bucket by the SARC door.
6. Paperwork that does not contain patient information may be disposed of in regular trash containers or paper recycle containers.
7. Paperwork or unused labels with patient information should be disposed of in the shredding containers.

### **Limitations of procedure**

1. Becton-Dickinson blood collection tubes are designed for skin puncture blood collections.
2. Becton-Dickinson does not recommend re-centrifuging gel tubes once the barrier has formed. Re-centrifugation could cause cell lysis, resulting in the release of intracellular contents into the serum or plasma.
3. Chemistry values obtained from skin puncture plasma may differ from those obtained from skin puncture serum, venous plasma or venous serum.
4. Conditions of collection must be within specified range of acceptability to ensure proper mixing and test results (volume, etc).

### **References**

1. Tietz, Norbert W., ed., Textbook of Clinical Chemistry, W.B. Saunders Company, Philadelphia, PA. 1986. pp. 175-177.
2. Becton Dickinson, Vacutainer Systems circular, Becton Dickinson and Company, Franklin Lakes, New Jersey.
3. Becton Dickinson, Microtainer circular, Becton Dickinson and Company, Franklin Lakes, New Jersey.