## TITLE: Proper Collection & Preparation of Patient Samples

### PURPOSE

Collection and preparation of patient samples is an important aspect of coagulation testing. Failure to use proper venipuncture and centrifugation techniques will cause erroneous results.

### PERSONNEL

Medical Technologists, Technicians, Clinical Associates and Nursing Personnel

### SPECIMEN

For routine coagulation tests such as PT, PTT, Fibrinogen and D-Dimer, the blue top tube with Soduim Citrate anticoagulant is to be used in the following ratios:

**9** parts blood to**1** part anticoagulant

3.2% buffered sodium citrate solution (blue top plastic tubes)

**NOTE**: The anticoagulant concentration in the plasma will be altered in samples from patients with a very high hematocrit. If the hematocrit is above 55%, recalcification testing, such as PT and APTT's can be prolonged. (See section III of this procedure for instructions on how to handle these specimens).

Follow Standard Precautions

### STEPWISE PROCEDURE

I. Collection of the blood sample

A. Tubes must be labeled with Soft ID collection labels, using positive patient ID

a. Name

b. Date & time

c. Room number

d. Initials of person collecting specimen

B. A clean trauma-free venipuncture should be obtained to reduce the contamination of the blood sample by tissue thromboplastin.

1. The tube should not be removed from the needle until the vacuum has drawn the full amount of blood specimen. The 3 ml size tube must be filled completely.

**\*Tubes have a minimum/maximum fill indicator line on them, the blood must fall within the indicator. Check the Example card for under/over filled amounts.**

D. After collection, the tube should be inverted gently several times to ensure adequate mixing of the blood and anticoagulant.

E. The blood must be sent to the lab immediately after drawing.

1. **CLSI (NCCLS)** continues to recommend that blood specimens for coagulation

testing be collected by venipuncture using a blood collection system that collects

the specimen directly into a tube containing the anticoagulant. Collection of the

blood through lines that have been previously flushed with heparin should be

avoided, if possible. If the blood sample must be drawn through an indwelling

catheter, possible heparin contamination and specimen dilution should be

considered. In this case, the line should be flushed with 5 mL of saline and then

the first 5mL of blood or 6 times the line volume must be discarded. For those

samples collected from a normal saline lock (capped off venous port) twice the

dead space volume of the catheter and extension set should be discarded.

(Peripheral draws are preferred).

If multiple specimens are collected, the coagulation specimen should preferably be collected in the first tube, with no discard tube prior, unless blood cultures are to be drawn. If using a butterfly needle, a discard non-additive tube is needed to draw blood through the empty space in the tubing and then a blue top can be used second. Any EDTA or heparin tubes should be collected after the citrate tube for coagulation.

II. Centrifugation of the blood sample

A. Visually inspect the test tube for clots and quantity of specimen before

centrifugation. Check tubes with applicator sticks prior to spinning.

B. Centrifugation of the whole blood specimen *should* be done within 30 minutes of collection.

1. Spin the specimen, capped, for a minimum of 10 minutes at a 4500 RPM.

Speed in RPM’s may vary to attain platelet poor plasma (<10 x 109/L).

D. The centrifuged plasmas should be tested immediately. If

delays are expected, **store samples at room temperature; 24 hours for PT’s**

**and run no more than** **4 hours for PTT’s, Fibrinogen’s and D-Dimer’s.**  **Protimes can be tested up to 24 hours post draw time at room temperature.**

Samples for unfractionated heparin testing should be centrifuged within one hour from the time of collection.

\*If testing cannot be performed within the above time frames, platelet poor plasma should be removed from the cells and frozen at -20°C for up to two weeks or at -70°C for up to 6 months.\*

E. Plasmas that are:

1. **Lipemic** - can be run on the CA-660, as the instrument adjusts somewhat, For very lipemic samples, the instrument will give you an error and no results. The test will have to be rejected, recollected with a comment for documentation and called to the floor.
2. **Icteric** - can be run on the CA-660, if there is an error, follow the protocol for errors in the quick guide.

3. **Hemolyzed** (mod-grossly hemolyzed) should **not** be used. Have the specimen rejected and recollected. \*Slightly hemolyzed specimens can be run with the canned message attached that “results may be affected”.

III. Unacceptable blood specimens to be rejected are:

A. Those not labeled properly.

B. Clotted specimens: Clots or fibrin strands found.

1. Improper amount of blood drawn in the tube.
2. Whole blood specimens transported on ice. Ice is not recommended due to

possible cold activation of factor VII, loss of von Willebrand factor and platelet

disruption. **Samples should be transported at room temperature.**

E. Samples with visually high (greater than 55) hematocrit need to be redrawn with the appropriate amount of anticoagulant. This can lead to prolonged PT and PTT results. Follow instructions below:

1. Use blue top tubes from the LCC area.

2. Remove all anticoagulant from blue top tubes and place in

red top vacutainer tube.

3. Dispense appropriate amount of anticoagulant in dry red tube

(see following chart).

4. Draw patient using a syringe and add appropriate amount of

whole blood to the tube with anticoagulant.

5. Mix well.

6. Process blood according to section II of this procedure.

CITRATE

HCT ANTICOAGULANT SAMPLE

\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_

100-82 0.1 4.9

81-71 0.2 4.8

70-55 0.3 4.7

20-10 0.7 4.3

10-1 0.8 4.2

F. Those samples accompanied with incomplete patient information. Information

required on the label:

1. Name of patient

2. Location of patient

3. Time and date drawn

4. Initials of person who drew the sample.

Contact the floor or the person drawing the sample with the

missing information. Mislabeled or unlabeled specimens are

to be automatically rejected. An Occurrence form is to be filled out in

accordance to the hospital “Red Rule” protocol.

1. Those specimens for PTT’s not sent to the lab within 4 hours after

collection. These specimens can be run, but a disclaimer or notation must be

indicated on the report.

### REFERENCES

1. Greiner bio-one vacuette product collection tips for coagulation testing, 0735005R1.

2. CLSI (NCCLS) standards for Collection, Transport, and Processing of Blood Specimens for Testing Plasma-Based Coagulation Assays and Molecular Hemostasis Assays; Approved Guideline- Fifth Edition; H21-A5. Vol. 28 No. 5 Wayne, PA: CLSI January 2008.

3. Siemens Healthcare Diagnostics Hemostasis Installation/Traing Services: Siemens

Installation Package Rev. 3.0.

4. Siemens Reagent Product Insert.