Specimen Collection – Venipuncture Procedure

I. Collecting and Processing of Specimens
   A. Blood
      1. Venipuncture Procedure (arm + dorsal/hand) – vacutainer, syringe, butterfly
         a. Approaching the Patient
            Correct patient identification
            Wash hands
            Have patient recite his/her name
            Wrist identification – mandatory
            must match requisition
            check ankle on babies and pediatrics
            Out-patients – ask patient to spell name
            In-patients – see wrist ID
            Unconscious patient – see wrist ID
            Unidentified patient –
               emergency – use temporary I.D. band
               non-emergency – wait for I.D.

            Explanation and Reassurance –
            Inspire confidence
            Conversation

            Ensure that patient has complied with test requirements, such as fasting (only water), NPO (nothing per oral), etc.

            Check for any allergies, such as to latex, adhesive bandages, etc.

         b. Positions
            Positioning the patient
            Vein accessibility
            Sitting vs. lying down

            Phlebotomist position – always in front in case of fainting.

         c. Applying the Tourniquet
            See previous lecture
d. **Veins Used - Antecubital Fossa**

Cephalic  
Median Cephalic  
Median  
Basilic  
Median Cubital Vein – vein of choice, anchored best  
Other Structures – avoid  
  Brachial artery – apply pressure 5 minutes  
  Cutaneous nerve – very painful  
  Tendon for the biceps muscle – always draw below crease

e. **Other Vein Sites**

Wrist (never palm side)  
Hand  
Ankle  
Foot

f. **Preparing Equipment**

Syringes  
  Assembly – always work plunger before procedure.  
  Plunger position – must be fully compressed  
Evacuated tube system  
  Needle  
  Safety needle guard  
  Safety holder  
  Vacuum tube position  
  Preventing blood leakage  
Butterfly  
  Appropriate size  
  too small – hemolysis  
  too big – rupture vein  
  Adaptor – syringe  
  tubes

g. **Selecting the Vein**

Apply tourniquet  
  Palpate area  
  Patient clenches fist – no pumping
Both arms – check if nothing in first arm
Application time – less than 1 minute

Vasodilation
Tourniquet
Fist clenched
Lower arm – hang down
Warm towel – arteriolization

Differentiating veins from arteries
Arteries – pulse
Veins – bounce back
  Sclerosed – hard
  “elastic tubes”

h. Cleansing the Puncture Site

Use 70% isopropyl alcohol wipe
  Circular motion, starting at the inside of the venipuncture site and working outward
Let air dry 30-60 seconds –
  Do not blow (or fan) on site!
  Do not wipe with cotton/gauze
Re-disinfect, if necessary

Caution: If vein needs to re-palpate the vein, wipe tip of gloved finger first with alcohol, and palpate above the point of needle insertion

* Blood cultures – special iodine scrub procedure
* Alcohol levels – Do not use alcohol to cleanse site!

i. Performing the Venipuncture (Right-handed Person)

Re-cleanse puncture site, if necessary
Inspect needle
Hold syringe or vacutainer in right hand; thumb on top, fingers underneath
Point needle in parallel direction to the vein
  Anchor (plant) vein
  Left thumb one inch below puncture site
  Left index-finger above site
  Left thumb and index finger – stretch skin
Introduce needle into vein; bevel up, at 15-30° angle
Back of fingers of right hand should rest on patient's arm as an anchor while needle enters vein. May have small drop of blood on top of skin. This sometimes
occurs with large veins that are close to skin. Do NOT change this hand; always have it be an anchor.

*Syringe* – pull back gently on the plunger and release to allow blood to fill syringe. Do not alter the position of the needle in the vein.

*Evacuated tube system* – push vacutainer tube into holder. Place first and second fingers of left hand against the top of the base of the holder and the thumb against the bottom of the tube. Hold tightly to holder to prevent movement. Do not push holder – may force needle through the vein.

Release tourniquet – when last tube is ½ filled.

Cover puncture site with dry cotton or gauze (do not press) and quickly withdraw the needle from the vein. Immediately apply pressure to the puncture site. Activate safety device on needle.

Instruct patient to apply pressure to the area, preferably with arm held above head for 2-3 minutes, or apply pressure yourself if necessary. **DO NOT BEND ARM.**

Apply pressure bandage –

Fold 2” gauze into quarters, place over wound, and apply bandage tightly over gauze.

If artery is accidentally stuck (as indicated by bright red blood the spurs into tube), phlebotomist applies pressure for 10 minutes.

j. **Transferring Blood - Syringe to Collection Tube**

Puncture diaphragm of stopper; **DO NOT PUSH ONTO PLUNGER**
Allow blood to run gently down the side of the tube
Invert gently – 10 to 12 times (if tube contains anticoagulant)

Alternate method – use BD blood transfer device

k. **Identifying Specimen**

Label *after* collection

- Patient’s first and last names (printed)
- Patient’s ID number
Name or initials of phlebotomist
Date of blood collection
Time of blood collection

**DO NOT PRE-LABEL TUBES**
If you don’t get the specimen, then the tubes are wasted
Computer printed labels are to be applied after obtaining specimens.

l. **Disposing of Used Equipment**

Paper, plastic wrappers - discard in waste basket in patient's room
Discard used needles and syringes discard in special sharps disposal containers
**NEVER RECAP!**
Discard contaminated gauze or cotton in biohazard waste container
Remove gloves, and **WASH HANDS!!**

m. **When leaving**

Thank the patient for his/her cooperation
Check to be sure that no items are left on the bed or table
Do not adjust the bed, if asked to do so; just let the nurse know of the patient’s request
Leave the room as it was found (bed and bed rails)

n. **Collecting of Multiple Samples**

While holding vacutainer holder with right hand and anchoring the back of the fingers on the patient’s arm, switch vacutainer tubes with left hand

**DO NOT ATTEMPT TO SWITCH HANDS ON THE HOLDER!**
It presents a risk of injury to patient, or movement of needle out of vein

To help make the removal to tubes easier, twist the tube as its being taken out of the holder

*Order of draw* (CLSI/NCCLS, per Strasinger) –
1. Sterile specimens (yellow stopper, blood culture)
2. Glass red stopper (plain, non-additive)
3. Light blue stopper (Na citrate)
4. Plastic red stopper (clot activator)
5. Red/gray, gold stopper (serum separator)
6. Green stopper (heparin)
7. Light green stopper (plasma separator)
8. Lavender or Pink stopper (EDTA)
9. Gray stopper (oxalate/fluoride)
10. Yellow/gray or orange stopper (thrombin clot activator)

**DO NOT DEVIATE FROM THIS ORDER**

If affiliates do different, follow this order anyway

**NOTE:** Handling of Routine Specimens

All tubes should be gently inverted 8-10 to ten times as soon as they are drawn, especially plastic red top

Vigorous mixing may cause hemolysis and should be avoided

Potassium, magnesium, and certain enzyme tests are examples of tests that cannot be performed on hemolyzed specimens

Inadequate mixing of gel separation tubes may prevent the additive from functioning properly and clotting may be incomplete

**Common Tests Affected by Additive Contamination**

<table>
<thead>
<tr>
<th>Contaminating Additive</th>
<th>Tests Potentially Affected</th>
</tr>
</thead>
<tbody>
<tr>
<td>Citrate</td>
<td>Alkaline phosphatase</td>
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<tr>
<td></td>
<td>Calcium</td>
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<tr>
<td></td>
<td>Phosphorus</td>
</tr>
<tr>
<td>EDTA</td>
<td>Alkaline phosphatase</td>
</tr>
<tr>
<td></td>
<td>Calcium</td>
</tr>
<tr>
<td></td>
<td>Creatine kinase</td>
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<tr>
<td></td>
<td>Partial thromboplastin</td>
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<tr>
<td></td>
<td>Potassium</td>
</tr>
<tr>
<td></td>
<td>Protime</td>
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<tr>
<td></td>
<td>Serum iron</td>
</tr>
<tr>
<td></td>
<td>Sodium</td>
</tr>
<tr>
<td>Heparin (all formulations)</td>
<td>Activated clotting time</td>
</tr>
<tr>
<td></td>
<td>Acid phosphatase</td>
</tr>
<tr>
<td></td>
<td>Calcium (some test methods)</td>
</tr>
<tr>
<td></td>
<td>Partial thromboplastin</td>
</tr>
<tr>
<td></td>
<td>Protime</td>
</tr>
<tr>
<td></td>
<td>Sodium (sodium formulations)</td>
</tr>
<tr>
<td>----------</td>
<td>-----------------------------</td>
</tr>
<tr>
<td>Oxalates</td>
<td>Acid phosphatase</td>
</tr>
<tr>
<td></td>
<td>Alkaline phosphatase</td>
</tr>
<tr>
<td></td>
<td>Amylase</td>
</tr>
<tr>
<td></td>
<td>Calcium</td>
</tr>
<tr>
<td></td>
<td>Lactate dehydrogenase</td>
</tr>
<tr>
<td></td>
<td>Partial thromboplastin</td>
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<tr>
<td></td>
<td>Potassium</td>
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<tr>
<td></td>
<td>Protime</td>
</tr>
<tr>
<td></td>
<td>Red cell morphology</td>
</tr>
<tr>
<td>Silica (clot activator)</td>
<td>Partial thromboplastin time</td>
</tr>
<tr>
<td></td>
<td>Protime</td>
</tr>
<tr>
<td>Sodium fluoride</td>
<td>Sodium</td>
</tr>
<tr>
<td></td>
<td>Urea nitrogen</td>
</tr>
</tbody>
</table>

(from McCall R, Tankersley C. Phlebotomy Essentials. 4th ed. Baltimore, Md.: Lippincott Williams & Wilkins. 2008.)

**RATIONALE FOR COLLECTION ORDER:**

<table>
<thead>
<tr>
<th>Order of Draw</th>
<th>Tube Stopper Color</th>
<th>Rationale for Collection Order</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blood cultures (sterile collections)</td>
<td>Yellow SPS</td>
<td>Minimizes chance of microbial contamination</td>
</tr>
<tr>
<td></td>
<td>Sterile media bottle</td>
<td></td>
</tr>
<tr>
<td>Glass non-additive tubes</td>
<td>Red</td>
<td>Prevents contamination by additives in other tubes</td>
</tr>
<tr>
<td>Coagulation tubes</td>
<td>Light blue</td>
<td>The first additive tube in the order because all other additives affect coagulation tests</td>
</tr>
<tr>
<td>Plastic clot activator tubes</td>
<td>Red</td>
<td></td>
</tr>
<tr>
<td>Serum separator tubes (SSTs)</td>
<td>Red and gray rubber</td>
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</tr>
<tr>
<td></td>
<td>Gold plastic</td>
<td></td>
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<tr>
<td></td>
<td></td>
<td>Filled after coagulation tests because silica particles activate clotting and affect coagulation</td>
</tr>
<tr>
<td></td>
<td></td>
<td>tests (carry-over of silica into subsequent tubes can be overridden by anticoagulant in them)</td>
</tr>
<tr>
<td>Plasma separator tubes (PSTs)</td>
<td>Green and gray rubber</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Light-green plastic</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Heparin affects coagulation tests and interferes in collection of serum</td>
</tr>
<tr>
<td>Additive</td>
<td>Color</td>
<td>Interference</td>
</tr>
<tr>
<td>------------------</td>
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<td>------------------------------------------------------------------------------</td>
</tr>
<tr>
<td>Green</td>
<td>specimens</td>
<td>causes the least interference in tests other than coagulation tests</td>
</tr>
<tr>
<td>EDTA tubes</td>
<td>Lavender Pink</td>
<td>Responsible for more carry-over problems than any other additive: elevates Na⁺ and K⁺ levels, chelates and decreases calcium and iron levels, elevates PT and PTT results</td>
</tr>
<tr>
<td>Plasma preparation tubes (PPTs)</td>
<td>Pink Pearl top</td>
<td></td>
</tr>
<tr>
<td>Oxalate/fluoride tubes</td>
<td>Gray</td>
<td>Sodium fluoride and potassium oxalate affect sodium and potassium levels, respectively, after hematology tubes because oxalate damages cell membranes and causes abnormal RBC morphology. Oxalate interferes in enzyme reactions.</td>
</tr>
</tbody>
</table>


This ‘Order of Draw’ has been set forth by CLSI (Clinical and Laboratory Standards Institute), and is the standard for best practice in the laboratory.

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