Specimen Collection – Phlebotomy Equipment and the Circulatory System

I. Collecting and Processing of Specimens
   A. Blood
      1. Venipuncture Equipment (including vacuum additives, etc.)
         a. Tourniquet

            **Purpose of use** - apply enough pressure to the arm to slow return of blood to the heart. This will dilate veins, which makes them more prominent and easier to find.

            **Types available** -
            - penrose drain (thin rubber tube used as surgical drain device)
            - strap or belt
            - rubber hose
            - blood pressure cuff
            - velcro fastener buckle release

            **Effects on laboratory specimen testing** -
            - too tight (block flow)
            - too long (hemoconcentration)
            - false elevated test results - such as, cholesterol, Iron (Fe), lipids, proteins, Potassium (K+), and enzymes

            **Sizes** -
            - adult pediatric
            - pediatric

            **Applying the tourniquet** -
            Will be demonstrated and performed in lab.

         b. Syringes

            **2 main parts**
            - barrel
            - plunger

            **Safety Shield**
            - Composition material - glass, or plastic
            - Needle attachment

            **Uses** -
            - blood culture collection
            - difficult veins
            - implanted port/CVC lines
            - blood gas analysis
Sizes -

1 cc to 100 cc (1 cc = 1 mL); cc is old term

c. Needles

Components

- bevel
- shaft
- hub
- length
- lumen

Sizes

diameter of lumen = gauge size
length = inches
common size = 21g 1 in

Safety devices

- sharps disposal containers
- eclipse needle guard
- pronto quick release holder
- Proguard needle holder

d. Butterfly/IV infusion set

Common sizes

- 21g, 23g, and 25g

Uses

- difficult veins
- infants and children

Safety devices

- BD Safety Lock
- angel wing butterfly

Can use with either syringe or specially-designed evacuated tube holder

e. Vacuum tubes

Components -

- vacuum stopper tubes
- needle - multisample

Holder -

- screw lock
- pronto quick release
- other disposable safety devices
B. Tubes

**Blood Cultures:**

**Yellow - Blood Cultures (SPS) –**

**Specimen Provided: Cells + Plasma in Broth**

a. yellow top - contains sodium polyanetholesulfonate as a preservative and anticoagulant
b. broth with resin, drawn in sets of two
   - aerobic - with exposure to air during incubation
   - anaerobic - without exposure to air during incubation

*When placing syringe into anaerobic culture, caution must be used to avoid possible injection of air into bottle. *

Resin in tubes absorbs antibiotics that may inhibit growth

Requires special scrub - sterile technique

a. green soap if allergic to alcohol or iodine
b. alcohol
c. iodine

**Anticoagulants:**

**EDTA (Lavender or Pink) ethylenediamine tetra-acetic acid**

**Specimen Provided: Whole Blood = Cells + Plasma**

Preferred for hematological studies

Prevents clotting by binding (chelate) calcium

Prevents platelet aggregation

Minimal distortion of white blood cells; excellent for preparation of blood smears

False low hct - shortdraw (excess EDTA)

False low WBC and platelet count (clotted - excess blood)

Hemolysis + cell distraction - (excess EDTA)

**Sodium Citrate (Blue or light blue)**

**Specimen Provided: Plasma**

Prevents clotting by binding (chelate) calcium

Preferred for coagulation studies

For valid results, a 1:9 ratio must be employed – “Full tube”

Short-draw results in prolonged clotting time (false positive result)

Specimen must be refrigerated if testing prolonged

False low westergren sedimentation rate due to incorrect ratio of citrate to specimen

**Heparin (Green) - 3 types**

**Specimen Provided: Plasma**

Prevents clotting by inactivation of thrombin and thromboplastin

Preferred for assays such as ammonia and plasma hemoglobin (Chemistry Department)
Not suitable for coagulation or hematological studies due to background staining on blood smears and its anti-thrombin properties

Lithium heparin - not used in lithium determinations
Sodium heparin - not used for electrolyte evaluations = elevated sodium levels
Ammonium heparin - not used for ammonia or blood urea nitrogen (BUN) levels

Oxalate (Gray) 2 types
Specimen Provided: Plasma + Cells +
Preservative
1. Sodium fluoride/potassium oxalate or
2. Lithium iodoacetate/heparin
Prevents clotting by binding (chelate) calcium
Heparin and potassium oxalate are anticoagulants
Not suitable for blood smears due to cellular morphology distortion
Antiglycolytic agent sodium fluoride or iodoacetate used for glucose and alcohol testing
Not suitable for enzyme determinations since fluoride destroys enzymes

Preservatives:
Sodium Fluoride (Gray)
Glycolytic Inhibition - prevents action of enzymes in red blood cells which metabolize glucose and alcohol
Oxalate binds calcium and prevents clotting
Primary use - glucose and alcohol levels
Not a suitable specimen for enzyme determinations since fluoride acts as an enzyme inhibitor
Not suitable for testing procedures which use enzyme reagents

Iodoacetate (Gray) (iodoacetic acid)
Glycolytic Inhibition - stabilizes glucose levels for 24 hours at room temperature
Usually added along with heparin as an anticoagulant

Thymol
Antibacterial agents
Usually added along with sodium fluoride

Non-Additive:
Specimen Provided: Serum
Plain Uncoated Clot Tube (Red Tube)
Primary use - blood bank (also chemistry and serology - serum use)
Tube of choice for blood bank - avoids interference with visual agglutination (silica)
No possibility of coating red cell membranes - optimum reactivity of antigenic sites
Other Additives:

Specimen Provided: Serum

1. Inert polymer barrier (SST tube) (red/gray marble or gold)
   - Gel is used to separate serum/cells after centrifugation
   - Not suitable for blood banking procedures due to coating of RBC membranes
   - Preferred by chemistry

2. Siliceous Earth (red plastic)
   - Clot activator - provides more surface area

3. Thrombin (orange)
   - Clot activator
   - Not a suitable specimen for coagulation studies
   - Primary use - STAT procedures in Chemistry
   - Must have in plain red plastic tubes to facilitate clotting

4. Silicon (CAT Tube = Red/Yellow)
   - Clot activator - provides more surface area
   - Decreases possibility of hemolysis
   - Prevents cell hang up on tube wall
   - Primary use - STAT procedures in chemistry
   - Can possibly bind some drugs
   - *May be used for emergency blood bank orders when blood products are needed STAT

Other Preservatives: (not vacuum tubes, used for specialty testing)

ACD solution (Acid Citrate Dextrose)
- Extends metabolism and life span of RBCs
- Used in Blood Bank - donor units

CPDA-1 (Citrate Phosphate Dextrose with Adenine)
- Extends metabolism and life span of RBCs - can be stored up to 35 days at 1-6°C

Alsever’s Solution
- RBC preservative
- Extends life span of RBCs
- Used in Blood Banking procedures

CPD Solution (Citrate Phosphate Dextrose)
- Extends metabolism and life span of RBCs - can be stored up to 21 days at 1-6°C
### SPECIMEN TYPE AND COLLECTION VACUUM TUBES

<table>
<thead>
<tr>
<th>Specimen Type</th>
<th>Collection Tubes (Stopper Color/Type)</th>
<th>Additive</th>
<th>Department</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blood/serum</td>
<td>Gray/red, gold</td>
<td>Inert polymer barrier + clot activator</td>
<td>Chemistry</td>
</tr>
<tr>
<td>Yellow and red</td>
<td>Inert polymer barrier</td>
<td></td>
<td>Chemistry</td>
</tr>
<tr>
<td>Yellow/grey, orange</td>
<td>thrombin</td>
<td></td>
<td>Chemistry</td>
</tr>
<tr>
<td>Red (glass)</td>
<td>None</td>
<td></td>
<td>Chemistry, Serology</td>
</tr>
<tr>
<td>Red (plastic)</td>
<td>Clot activator</td>
<td></td>
<td>Chemistry, Serology</td>
</tr>
<tr>
<td>Whole Blood, plasma</td>
<td>Lt green or green/gray</td>
<td>Inert polymer barrier (gel) + Li heparin - pl</td>
<td>Chemistry</td>
</tr>
<tr>
<td>Light green</td>
<td>Li or Na Heparin - wb, pl</td>
<td></td>
<td>Chemistry</td>
</tr>
<tr>
<td>Light blue</td>
<td>3.2 or 3.8 % Na Citrate</td>
<td></td>
<td>Coagulation</td>
</tr>
<tr>
<td>Lavender</td>
<td>K&lt;sub&gt;3&lt;/sub&gt; EDTA (glass) - wb</td>
<td></td>
<td>Hematology</td>
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<tr>
<td></td>
<td>K&lt;sub&gt;2&lt;/sub&gt; EDTA (plastic) - wb</td>
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<tr>
<td>Black</td>
<td>Na citrate - wb</td>
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<td>Hem - sed rate</td>
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<tr>
<td>Pink</td>
<td>EDTA - wb, pl</td>
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<td>Blood Bank</td>
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<tr>
<td>White</td>
<td>EDTA + gel - pl</td>
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<td>Molecular Diagnostics</td>
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<tr>
<td>Gray</td>
<td>Na fluoride+K oxalate - pl</td>
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<td>Chemistry – glucose, lactate, alcohol</td>
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<tr>
<td></td>
<td>Na fluoride - s</td>
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<tr>
<td></td>
<td>Na fluoride+K&lt;sub&gt;2&lt;/sub&gt; EDTA - pl</td>
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<tr>
<td></td>
<td>Li iodoacetate - s</td>
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</tr>
<tr>
<td></td>
<td>Li heparin+iodoacetate - pl</td>
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<tr>
<td>Blood/serum</td>
<td>Royal blue</td>
<td>Sodium (Na) Heparin - pl</td>
<td>Chemistry - low level trace elements, toxicology, nutrients</td>
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<td></td>
<td>Na&lt;sub&gt;2&lt;/sub&gt; EDTA - pl</td>
<td>None - s</td>
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<tr>
<td>Tan</td>
<td>Sodium Heparin - pl</td>
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<td>Chemistry - lead</td>
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<tr>
<td></td>
<td>K&lt;sub&gt;2&lt;/sub&gt; EDTA (plastic) - pl</td>
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<td></td>
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<tr>
<td>Yellow</td>
<td>Na polyanetholesulfonate (SPS) - wb</td>
<td></td>
<td>Microbiology - blood cultures</td>
</tr>
</tbody>
</table>
C. Additional Equipment

a. Miscellaneous items
gloves
gauze pads - non sterile 2" x 2"
cotton balls - stick to wound
antiseptic
  70% isopropyl alcohol
  routine venipuncture
  iodine (Betadine)
  blood cultures
  arterial blood gases
  blood alcohol levels
  green soap
  patients allergic to iodine
  blood cultures
  poor personal hygiene
  bandages - pressure
  marking pen (water resistant)
  sharps disposal container - ridged plastic
  microscope slides
  capillary tubes

b. Dermal puncture equipment (more later)

Uses
burn patients
difficult veins
extreme obesity
infants (small volume)
bedside clinical testing

Components
lancets
capillary collecting devices
capillary pipette (tube)
plain
heparinized
capillary pipette sealer

Microtainers
non-additive
SST
EDTA
heparin
heparin/gel

**Controlled puncture device**
controls depth
determines type – slit, hole, etc.

**C. Bleeding time test equipment**
Blood pressure cuff (Sphygmomanometer)
Filter paper disks (Whatman’s #1)
Butterfly bandage
Incision Device
   - Simplate
   - Surgicutt

**Bleeding Time Procedure**
d. Request slips (computer generated or manual forms)

Pertinent information
- Patient’s full name
- Patient’s ID number
- Date of collection
- Time of collection
- Phlebotomist’s initials
- Patient’s room number or outpatient status

Test(s) ordered
- ex: CBC, Glucose etc.

Time - when test is to be collected
- AM/PM
- routine/daily
- peak/trough

II. Heart and Circulatory System
All body systems are linked by the cardiovascular system, a transport network that can affect every cell, tissue, and organ within seconds. The primary components of the cardiovascular system include blood (circulating fluid), the heart (a pump), and the circulatory system or blood vessels (numerous connected tubes).

The human heart is about the size of a man’s fist and is shaped like an upside-down pear with the tip, or apex, pointing to the left side of the body.

a. Pathway of blood through the body

The flow of blood through the heart is very orderly. Blood progresses through the heart to the lungs (where it receives oxygen), back to the heart, and then out to the body tissues and organs. The normal flow is as follows:

- The systemic circulatory system.
- The pulmonary circulatory system.

- The blood from the parts of the body, travel through the veins to the inferior/superior vena cava (vein) into the right atrium, through the tricuspid valve into the right ventricle.
- From there it is pumped through the pulmonary semilunar valve into the left/right pulmonary arteries to each lung.
- In the lungs the CO₂ in the capillaries is exchanged for O₂ in the lungs.
- The pulmonary veins then carry the oxygenated blood from the lungs to the left atrium of the heart.
- The blood flows through the mitral valve into the left ventricle that contract to pump the blood through the aortic semilunar valve into the aorta.
- Blood travels throughout the body via the arteries to the capillaries in the tissues where the $O_2$ is exchanged for $CO_2$ waste in the tissues.
- Blood then travels to the vena cava with help of valves and the process continues.

**b. Blood (erythrocytes, leukocytes, and thrombocytes)**

All blood cells develop from undifferentiated stem cells in the hematopoietic (blood-forming) tissues, such as the bone marrow. Circulating blood cells are classified as:

- red blood cells (RBCs, or erythrocytes)
- white blood cells (WBCs, or leukocytes)
- platelets (thrombocytes)

Within each mature RBC are millions of hemoglobin molecules.

There are 5 types of WBCs:

- Neutrophils (40-60%)
- Lymphocytes (20-40%)
- Monocytes (3-8%)
- Eosinophils (1-3%)
- Basophils (0-1%)
The percentage of whole blood that is occupied by red cells is called the hematocrit (Hct or crit), and it is an important and commonly ordered laboratory test.

The liquid portion of the blood is called plasma.

If a blood specimen is allowed to clot, the result is serum plus blood cells meshed in a fibrin clot.

c. Terms and functions of the blood vessels, heart, and blood

Three kinds of blood vessels exist in the human body: arteries, veins, and capillaries.

**Arteries** are highly oxygenated vessels that carry blood away from the heart (efferent vessels).

**Veins** carry the blood towards the heart (afferent vessels).

Phlebotomists should be familiar with the principle veins of the arms and legs. The forearm vein most commonly used for venipuncture is the **median cubital** vein because it is generally the largest and best-anchored vein. Others that are acceptable are the **basilic** vein, the **cephalic** vein.

Microscopic vessels that carry blood and link arterioles to venules (minute veins) are called **capillaries**.

**Hemostasis** (*not to be confused with homeostasis*) is the maintenance of circulating blood in the liquid state and retention of blood in the vascular system by preventing blood loss.

Disorders (some) of the hemostatic (coagulation) process:

- Disseminated intravascular coagulation (DIC) – spontaneous activation of the coagulation system by foreign substances, causing depletion of platelets and coagulation factors, etc.

- Hemophilia – hereditary disorder characterized by excessive bleeding because of lack of coagulation factor, usually factor VIII.

**Diagnostic assessment of cardiovascular system:**

Blood pressure and pulse rate measurements are performed as part of routine physical assessments.

The **pulse** is a measurement of the number of times the heart beats in a minute. Pulse rate occurs as a result of pressure when the ventricles contract and blood is forced through the arteries.

**Blood pressure**, which is measured using a sphygmomanometer or blood pressure cuff, is a measurement of the force or pressure being exerted by the blood on the walls of the artery. The first number is the systolic pressure,
the ventricular contraction. The second number is the diastolic pressure, during the relaxation of the ventricles.

d. Disorders of the heart, correlated lab tests, and medications

Vessels –
- Aneurysm - bulge in the wall that can burst, causing severe hemorrhage
- Arteriosclerosis - hardening of the artery walls, contributing to aneurysm or stroke
- Atherosclerosis - form of arteriosclerosis which has accumulation of lipids, etc., walls of arteries, causing the vessel lumen to narrow and stimulate clot formation
- Embolism - obstruction of blood vessel by moving blood clot or foreign matter in vascular system; tissue destruction or death occurs if embolus lodges in an organ
- Phlebitis - inflammation of the vein wall causing pain and tenderness
- Thrombosis - obstruction of a blood vessel by a stationary blood clot, causing and aching pain
- Varicose veins - swollen peripheral veins caused by damaged valves

Heart –
- Angina Pectoris - sharp chest pain caused by decreased blood flow to the heart, usually because of an obstruction in the coronary arteries
- Bacterial endocarditis - inflammation of the inner lining of the heart caused by a bacterial infection, usually strep
- Congestive heart failure - a chronic disorder, where the ability of the heart to pump blood efficiently is impaired, causing fluid accumulation in the lungs and other tissues
- Myocardial infarction - death (necrosis) of the heart muscle caused by a lack of oxygen to the myocardium because of an occluded coronary artery, commonly known as a heart attack
- Symptoms include pressure in chest or down part of left arm, nausea, sweating, dizziness, shortness of breath, etc.
- Pericarditis - inflammation of the membrane surrounding the heart (pericardium), induced by bacteria, viruses, trauma, or malignancy
- Rheumatic heart disease - autoimmune disorder affecting heart tissue following a strep infection
### Correlation of diagnostic tests and clinical conditions:

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<th>Clinical Correlations</th>
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<td>Heparin therapy</td>
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<td>APTT (Activated partial thromboplastin time)</td>
<td>Heparin therapy or coag. disorder</td>
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<tr>
<td>angiogram</td>
<td>Blood vessel integrity</td>
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<td>Antibody (ab) screen</td>
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<td>ASO (antistreptolysin O) titer</td>
<td>Rheumatic fever</td>
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<tr>
<td>Antithrombin III</td>
<td>Coagulation disorders</td>
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<tr>
<td>Apo-A, Apo-B lipoprotein</td>
<td>Cardiac risk</td>
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<td>AST (Aspartate aminotransferase)</td>
<td>Cardiac muscle damage</td>
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<tr>
<td>Bilirubin</td>
<td>Hemolytic disorders (+ liver function)</td>
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<tr>
<td>Bleeding time</td>
<td>Platelet function</td>
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<tr>
<td>Blood culture</td>
<td>Microbial infections</td>
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<tr>
<td>Blood group &amp; type</td>
<td>ABO group, type, Rh factor</td>
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<tr>
<td>Bone marrow</td>
<td>Blood cell disorders</td>
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<tr>
<td>BNP (Brain natriuretic peptide)</td>
<td>Congestive disorders</td>
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<td>CRP (C-reactive protein)</td>
<td>Inflammatory disorders</td>
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<td>Cardiac catheterization</td>
<td>Coronary artery examination</td>
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<tr>
<td>Cholesterol</td>
<td>Coronary artery disease</td>
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<td>CBC (complete blood count)</td>
<td>Bleeding disorders, anemia, or leukemia</td>
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<tr>
<td>CT scan (Computerized axial tomography)</td>
<td>soft-tissue examination</td>
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<tr>
<td>CK (Creatine kinase)</td>
<td>Myocardial infarction</td>
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<tr>
<td>CK-MB (Creatine kinase heart isoenzyme)</td>
<td>Myocardial infarction</td>
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<tr>
<td>DAT (Direct antihuman globulin test)</td>
<td>Anemia, or hemolytic disease of the newborn</td>
</tr>
<tr>
<td>Echocardiogram</td>
<td>Cardiac abnormalities</td>
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<tr>
<td>ECG (Electrocardiogram)</td>
<td>Myocardial damage</td>
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<td>ESR (Erythrocyte sedimentation rate)</td>
<td>Inflammatory disorders</td>
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<td>FDP (fibrin degration products)</td>
<td>Disseminated intravascular coagulation</td>
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<td>Fibrinogen</td>
<td>Coagulation disorders</td>
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<tr>
<td>Hct (hematocrit)</td>
<td>Anemia</td>
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<td>Hb/Hgb (hemoglobin)</td>
<td>Anemia</td>
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<td>Hemoglobin electrophoreses</td>
<td>Hemoglobin abnormalities</td>
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<td>HDL (high-density lipoprotein)</td>
<td>Coronary risk</td>
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<tr>
<td>Fe (iron)</td>
<td>Anemia</td>
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<td>LD (lactate dehydrogenase)</td>
<td>Myocardial infarction</td>
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<tr>
<td>LDL (low-density lipoprotein)</td>
<td>Coronary risk</td>
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<td>Myoglobin</td>
<td>Anemia</td>
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<td>Plt (platelet count)</td>
<td>Bleeding tendencies</td>
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<td>PT (prothrombin time)</td>
<td>Coumadin therapy, coagulation disorders</td>
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<td>Retic (reticulocyte count)</td>
<td>Bone marrow function</td>
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<tr>
<td>Sickle cell screening</td>
<td>Sickle cell anemia</td>
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<td>Stress test</td>
<td>Cardiac function</td>
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<td>TIBC (Total iron biding capacity)</td>
<td>Anemia</td>
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<td>Triglycerides</td>
<td>Coronary artery disease</td>
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<tr>
<td>Troponin I and T</td>
<td>Myocardial infarction</td>
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</table>
f. **Type of medication, names, and purposes:**

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<thead>
<tr>
<th>Type</th>
<th>Generic and Trade Names</th>
<th>Purpose</th>
</tr>
</thead>
<tbody>
<tr>
<td>ACE inhibitors</td>
<td>Accupril, Capoten, Lotensin, Vasotec, Zestril</td>
<td>Lower blood pressure</td>
</tr>
<tr>
<td>Angiotensin receptor blockers</td>
<td>Avapro, Cozaar, Diovan</td>
<td>Lower blood pressure</td>
</tr>
<tr>
<td>Antiarrhythmic agents</td>
<td>Betapace, Cordarone, Inderal, Lanoxin, Mexitil, Norpace, Qinidx, Rythmol, Tambcor, Tonocard</td>
<td>Treat cardiac arrhythmias</td>
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<tr>
<td>Anticoagulants</td>
<td>Aggrenox, Aethrombin K, Coumadin, Heparin sodium, Lovenox, Plavix, Ticlid</td>
<td>Inhibit blood cloting</td>
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<td>Antihypertensive agents</td>
<td>Aldomet, Capoten, Cardizem, Catapres, Isoptin, Lopressor, Norvasc, Procardia, Cedilanid D, Crysstodigin</td>
<td>Lower blood pressure</td>
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<tr>
<td>Hematinic agents</td>
<td>Feosol, ferrous fumarate, ferrous gluconate, ferrous sulfate, Niferex, Trinsicon</td>
<td>Treat iron deficiency anemia</td>
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<tr>
<td>Hemostatic agents</td>
<td>Amicar, Humafac, Proplex, Protamine, Surgicel, vitamin K</td>
<td>Stop bleeding</td>
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<tr>
<td>Hypolipidemics</td>
<td>Atromid-S, Lescol, Lipitor, Lopid, Mevacor, niacin, Pravachol, Questran, TriCor, WelChol, Zocor</td>
<td>Lower lipid blood levels</td>
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<tr>
<td>Thromobolytic agents</td>
<td>Activase, Eminase, streptokinase, urokinase</td>
<td>Dissolve clots</td>
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<tr>
<td>Vasodilators</td>
<td>Apresoline, Cardilate, nitroglycerin, Sorbitrate</td>
<td>Lower blood pressure</td>
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<tr>
<td>Vasosuppressors</td>
<td>Aramine, Dopastat, levorphanol tartrate</td>
<td>Lower blood pressure</td>
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