Collection of clinical samples

Types of specimen samples collected for analyses

1. Blood-Plasma-Serum
2. Excretory products (Urine, Feces & Sweat)
3. Cerebrospinal fluid
4. Amnoitic fluid
5. GI tract juices & secretions (saliva, gastric juice, duodenal & jejunal secretions)
6. Pathological fluids (paracentesis- peritonial fluid & calculi)
7. Dietary intake samples

Factors affecting

1. Patient determined value
2. Technical Consideration

Patient determined values:

- Age & Sex
- Body size (weight/height)
- Physiological & mental states (pregnancy, menstrual cycles)
- Medication
- Ethnicity & domicile
- Time of the day
- Posture during test
- Diet & interval from last meal
- Exercise

Technical Consideration

- Site of collection (venous & capillary)
- Cleanliness
- Interval between collection and receipt of sample in laboratory
- Storage condition
- Methods (qualitative/quantitative/semiquantitative)

Parameters measured:

- Blood : Gases; Urea; Glucose
- Plasma & Serum: Proteins & Enzymes; Metabolites(cholesterol; uric acid); Electrolytes.
- Urine: Sugar; Proteins; Bile salts; Pigments; Steroids

Collection of blood

- Antecubital vein/prominent veins of fore arm
- Arterial blood (radial, brachial, femoral artery) – tip of fingers/ear lobe – children/multiple samples needed/veins not found.
- Anticoagulant needed
- **Plasma:** anticoagulants + centrifuged at 2000 rpm – supernatant
- **Serum:** glass container + kept for 30 mins-1hr+ centrifuged at 2000 rpm – supernatant

Precautions:

- **Venous occlusion/prolonged stasis:** hypoxia & variation in components (raised in plasma levels).
- **Hemolysis:** freezing & thawing should be avoided.

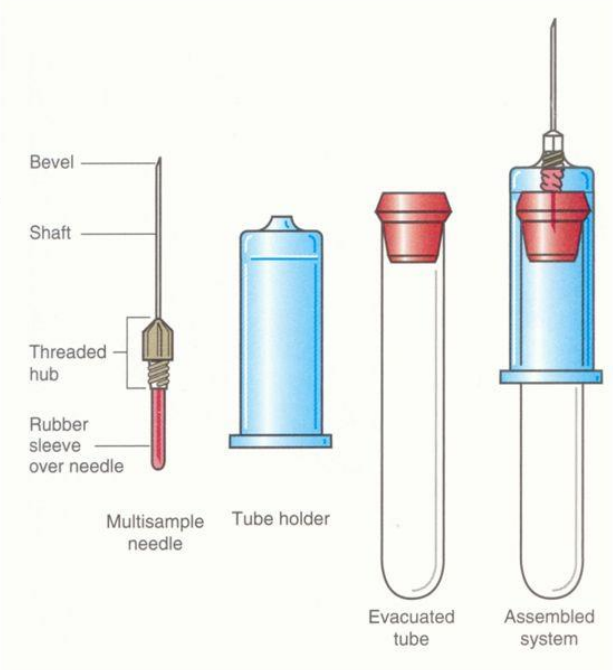
Anticoagulants:

Salts: precipitation of Ca

- EDTA, Oxalates, NaF, Heparin
- Concentrations (2mg/ml for EDTA/NaF; 0.2mg/ml for heparin etc.) – **Important**
- NaF: inhibits enolase of glyolysis (mixed as 1:3 with potassium oxalate)

Equipment:

8. Tube holder/ vacutainer adapter
   - Threaded
   - Flanges



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Vacutainers

Gel & clot activators:

1. Original character of blood
2. Hemolysis prevented
3. Silica gel – clotting
4. Barrier gel separates serum from fibrin & cells – prevents substance exchange.

Preservation & Storage of samples

- Sterility
- Freezing for longer storage (lyophilized)
- Addition of preservatives (to prevent chemical change & microbial growth like formaldehyde, thymol, toulene and chloroform)
- Serum is preferred over plasma.

Normal range = value found in persons with good health (95% of people)
depends on method chosen

Serum = blood – erythrocytes - fibrinogen
Plasma = blood – erythrocytes

Sensitivity = TP/(TP+FN)
Specificity = TN/(TN+FP)
Accuracy=(TN+TP)/(TN+TP+FN+FP)

Pre-analytical errors:

- Hemolysis (RBC)
- Lipemic (Lipids)
- Icteric (Bilirubin)
- Stale
- QNS (quantity not sufficient)
- PNC (proportion not correct)

Control = monitor status of analysis to maintain performance within desired limits

Calibrator = material/device with known qualitative & quantitative characteristics to standardize the method/instrument.

Methods for estimation

- Sensitivity (minute amount of analyte)
- Specificity (discriminating the analyte and the interfering compounds)
- Precision(scattered around mean-further measurements show similar results)
- Accuracy (tend/shift- conformity towards true value) – TP, TN, FP, FN
- Random error

Units & Reference values:

(1) Metabolites (glucose, urea) = mg/dl or mmol/L
(2) Electrolytes (sodium, potassium) = mmol/L
(3) Enzymes = units/L or U/L

Estimation of blood pressure using sphygmomanometer

Subject

- Position: supine, seated, standing.
- In seated position, the subject's arm should be flexed.
- The flexed elbow should be at the level of the heart.
- If the subject is anxious, wait a few minutes before taking the pressure.

Procedure

1. To begin blood pressure measurement, use a properly sized blood pressure cuff.
2. The length of the cuff's bladder should be at least equal to 80% of the circumference of the upper arm.
3. Wrap the cuff around the upper arm with the cuff's lower edge one inch above the **antecubital fossa.**
4. Lightly press the stethoscope's bell over the **brachial artery** just below the cuff's edge. Some health care workers have difficulty using the bell in the antecubital fossa, so we suggest using the bell or the diaphragm to measure the blood pressure.
5. Rapidly inflate the cuff to 180mmHg. Release air from the cuff at a moderate rate (3mm/sec).
6. Listen with the stethoscope and simultaneously observe the sphygmomanometer. **The first knocking sound (Korotkoff) is the subject's systolic pressure. When the knocking sound disappears, that is the diastolic pressure** (such as 120/80).
7. Record the pressure in both arms and note the difference. Record the subject's position (supine), which arm was used, and the cuff size (small, standard or large adult cuff).
8. If the subject's pressure is elevated, measure blood pressure two additional times, waiting a few minutes between measurements.

Precautions

•Aneroid and digital manometers may require periodic calibration.

•Use a larger cuff on obese or heavily muscled subjects.

•Use a smaller cuff for pediatric patients.

•For pediatric patients a lower blood pressure may indicate the presence of hypertension.

• Don't place the cuff over clothing.

• Flex and support the subject's arm.

In some patients the Korotkoff sounds disappear as the systolic pressure is bled down. After an interval, the Korotkoff sounds reappear. This interval is referred to as the "auscultatory gap." This pathophysiologic occurrence can lead to a marked under-estimation of systolic pressure if the cuff pressure is not elevated enough. It is for this reason that the rapid inflation of the blood pressure cuff to 180 mmHg was recommended above. The "auscultatory gap" is felt to be associated with **carotid atherosclerosis and a decrease in arterial compliance in patients with increased blood pressure.**