CHAPTER 19

CLINICAL LABORATORY

INTRODUCTION

A basic knowledge of clinical procedures is critical for the Hospital Corpsmen (HM), particularly those working at small dispensaries and isolated duty stations without the supervision of a Medical Officer. Clinical laboratory results aid health care providers to make accurate and timely diagnoses and treatment plans for their patients.

This chapter will outline laboratory administrative responsibilities, ethics in the laboratory, the microscope, blood collection techniques, the complete blood count, and urinalysis. Additional information includes a basic understanding of bacteriology, serology, operational readiness, and the Walking Blood Bank.

THE HOSPITAL CORPSMAN AND THE CLINICAL LABORATORY

LEARNING OBJECTIVE:

Explain clinical laboratory administrative procedures and ethics policy.

The HM is not expected to make a diagnosis from test findings or to institute definitive treatment based upon them. The availability of various communication methods aids the HM in giving a clearer clinical picture to the provider. This chapter is not intended to replace laboratory manuals, test procedures, or policies, but will provide basic laboratory principles.

Accuracy and attention to detail are essential to obtain optimum test results. These tests are only aids to diagnose a patient. Many other clinical factors must be taken into consideration before treatment may be started.

ADMINISTRATIVE PROCEDURES AND RESPONSIBILITIES

The ability to understand clinical laboratory tests is a commendable attribute of the HM. The entire testing effort will be wasted if proper documentation and filing practices are ignored, or the test results are misfiled. As a member of the medical team, it is the responsibility of the HM to make sure established administrative procedures are followed with regard to accurate patient and specimen identification. This includes ensuring laboratory reports are handled and filed properly in medical records.

Test results are a part of the patient's treatment record(s). Test results have a bearing upon the patient's immediate and future diagnosis and medical history.

Laboratory Request Forms

The Armed Forces have gone to great lengths to produce effective forms that serve a purpose with a minimum of confusion and chance for error. These forms are Standard Forms (SF) in the 500 series. With the exception of SF-545 (Laboratory Report Display), SF laboratory forms have been replaced by printed copies of laboratory results from computerized laboratory information systems (CHCS and AHLTA), or locally developed chits that meet the needs of each operational platform. Laboratory reports are filed on top of or attached to the SF-545 (see fig 19-1) located inside the patient's health record. For a complete listing of SF forms and their purposes, refer to the Manual of the Medical Department (MANMED), NAVMED P-117.

Laboratory request forms are not the only means by which healthcare providers can order laboratory tests. Many of today's treatment facilities have computerized laboratory systems.
These systems enable providers to enter laboratory test requests into computers located in their spaces. After providers enter their test requests, patients may report immediately to the Laboratory Department, where specimens are obtained and tests are performed.

**Use of Laboratory Request Forms**

Write information on the laboratory request forms in black or blue-black ink. Use a separate laboratory request form for each patient. Document the patient's full name, family member prefix and sponsor's social security number, rate/rank, date of birth, status, and branch of service in the "Patient Identification" block. Additionally, identify the ward or department ordering the test in this block. Computer-generated laboratory test requests require the same patient identification data as manual laboratory requests and the required information is automatically populated on the requests. HMs are required to verify the patient information before specimen collection and labeling.

The results of laboratory test are closely associated with the patient's health and treatment. The requesting provider's name must be included on the request. This practice ensures that reports/results get back to the requesting provider as soon as possible.

In addition to identifying the appropriate test on the form, enter any specific instructions in the area provided for remarks (e.g., "Clean catch midstream to rule out urinary tract infection"). Because the data requested, the date reported, and the time of specimen collection are required to support the clinical picture, information should be clearly written on the request.

**Patient and Specimen Identification**

Before accepting laboratory request forms and specimens in the laboratory, check patient identification information on both the request form and the specimen container label for completeness and legibility. Ensure the specimen(s) label(s) and request form information match with the patient. Proper documentation of patient identification on these items prevents errors.

**Filing Laboratory Forms**

After providers have reviewed laboratory test reports, they will initial or sign the form to indicate the review of the test results. This acknowledgement may be electronic via AHLTA or CHCS, or via initial/signature on a hard copy lab chit. After the provider releases the laboratory report, it will be filed in the patient's treatment record. Manual, automated, or computer-generated laboratory test reports will be placed above SF-545 (Fig. 19-1) in the health record. All forms will be filed chronologically with the each new result placed on top of the previous results.
Figure 19.1.—SF-545, Laboratory Report Display
ETHICS AND GOOD PRACTICES IN THE LABORATORY

LEARNING OBJECTIVES:

Identify the correct steps to perform blood collection by the finger puncture method and venipuncture method.

Explain Universal Precautions and other safety precautions that apply to blood collection.

The nature of laboratory tests and their results will be treated as a confidential matter between the patient, the provider, and the performing technician. Chapter 16 of the MANMED outlines the Navy’s ethics policy with regard to disclosure of the contents of a patient’s medical record, including lab reports. Other agencies that regulate medical ethics and patient confidentiality include The Joint Commission, Medical Inspector General (MED IG), and the Department of Health and Human Services (HHS). Of specific concern is the Health Insurance Portability and Accountability Act (HIPAA) enacted by Congress and enforced via HHS detailing how protected health information (PHI) may be transmitted and released. It is good practice to prevent unauthorized access to these reports, to leave interpretation of the test results to the requesting provider, and to refrain from discussing results with the patient. Always refer the patient back to their requesting provider for all laboratory results.

There are two principal methods of obtaining blood specimens: the capillary method and the venipuncture method. For most clinical laboratory tests requiring a blood specimen, venous blood obtained by venipuncture is preferred. Infection control practices, equipment requirements, and step-by-step instructions on performing both of these blood collection methods will be discussed in the following sections.

UNIVERSAL BLOODBORNE PATHOGEN PRECAUTIONS (See www.osha.gov and Standard 1910.1030 for complete details)

Under the concept of "Universal Bloodborne Pathogen Precautions" outlined by Occupational Safety & Health Administration (OSHA), all human blood and certain other human body fluids are treated as if known to be infectious for HIV, HCV, and other bloodborne pathogens, and therefore considered potentially infectious. Remember from Chapter 9 that Universal Precautions are a subset of Standard Precautions. The following Universal Precautions are in effect for all phlebotomy procedures:

- Gloves are required to be worn in conjunction with proper hand washing techniques
- Gloves will be disposed of after each patient
- Needles and other sharp instruments used in the blood collection process will be handled with extreme caution and disposed of in biohazard sharps containers
- Sharps containers will be conveniently located near phlebotomy work sites to reduce the distance between patient care and sharps disposal
- Absorbent materials, such as cotton 2 x 2's used to cover blood extraction sites, normally contain only a small amount of blood and can be disposed of as general waste
- If a large amount of blood is absorbed, the absorbent material will be placed in a biohazard waste container and treated as infectious waste
- Clean phlebotomy work site equipment and furniture daily with a disinfectant, or as needed after patient care
CAPILLARY BLOOD COLLECTION

Capillary blood collection is performed when a small quantity of blood is needed for testing as in the case of some pediatric blood draws. It may also be used when access to normal venipuncture draw sites are limited on a patient such as severely burned patients or ICU patients. Most adult capillary blood collections are from the finger. Capillary blood collections for newborns may occur from the heel.

Materials Required for Capillary Finger Puncture Procedure

To perform a finger puncture, the following materials are required:

- Sterile gauze pads (2" x 2")
- 70% isopropyl alcohol or povidone-iodine solution pads
- Blood lancets
- Plastic Capillary tubes
- Bandages

Arrange the equipment in an orderly manner and have it within easy reach. The HM will wash hands before and after each procedure.

Capillary Finger Puncture Procedure

To perform a finger puncture, follow the steps given below:

1. Explain the procedure to the patient.
2. Using the middle or ring finger, warm the site to make collection easier and faster. Warming the site reduces the tendency to squeeze the site.
3. Cleanse the fingertip with an alcohol pad or povidone-iodine solution and let dry.

4. Locate the correct puncture location on the finger. Always puncture away from the midline of the finger or heel to prevent injury to the bone (Fig 19-2). Do not puncture parallel to the grooves or lines of the fingerprint. A parallel puncture will allow blood to run down the finger rather than form a well rounded drop, and make collection difficult.

![Image](image-url)

Figure 19-2.—Vein puncture; A. Finger puncture; B. Heel Puncture


5. Take a lancet and make a quick stab no greater than 2 mm deep on the side of the finger (off-center). Commercially produced single-use lancets are available for ease of use and patient safety to control the puncture depth.
6. Wipe away the first drop of blood with a sterile 2 x 2 gauze. This prevents contamination of the specimen with excess tissue fluid. Avoid squeezing the fingertip to accelerate bleeding as this tends to dilute the blood with excess tissue fluid. Position the site downward to enhance blood flow and apply gentle intermittent pressure to tissue surrounding a finger puncture site.

7. Collect blood in the correct specimen container by “scooping” blood one drop at a time.

8. When the required amount of blood has been obtained, apply a pad of sterile gauze, instruct the patient to apply pressure, and then apply a bandage.

When dealing with newborns, infants, and very small children, the heel or great toe puncture may be used to obtain a blood specimen. This method is performed in a similar fashion. Additional training is required before these methods are performed by the HM.

**VENIPUNCTURE (VACUTAINER METHOD)**

Venipuncture is defined as the puncture of a vein for drawing blood. For the convenience of technician and patient, arm veins are best for obtaining a blood sample. If arm veins cannot be used due to interference from bandage or IV therapy, thrombosed or hardened veins, post-surgical requirements, etc., consult a supervisor for instructions on the use of hand or foot veins.

**Materials Required for Venipuncture**

To perform a venipuncture, the following materials are required:

- Sterile gauze pads (2" x 2")
- 70% isopropyl alcohol or povidone-iodine solution pads
- Tourniquet
- Vacutainer needles and holder with safety device to prevent accidental needle sticks
- Vacutainer tube(s) appropriate for the test to be performed

Arrange the equipment in an orderly manner and have it within easy reach. The HM will wash hands before and after the procedure.

**Venipuncture Procedure**

The patient must be positioned so that the vein is easily accessible and the HM is able to perform the venipuncture in a comfortable position. Always have the patient either lying in bed or sitting in a chair with the arm propped up.

**NOTE:**

Do not draw blood from an arm with IV fluid running into it or on the same side of a patient's mastectomy. Choose another site.

The IV fluid will alter test results and drawing blood from the same side of a patient's mastectomy can cause permanent damage.

If a patient has IV fluids running into both arms, consult a supervisor for instructions on the correct site for blood collection.

**WARNING:**

Never perform a venipuncture with the patient standing up.

If the patient faints, serious injury could result.

Safeguards should be in place to prevent patients from falling forward when they are seated.
To perform venipuncture, follow the steps given below.

1. Explain the procedure to the patient.

2. Apply tourniquet around the arm approximately 3 to 4 inches above the intended venipuncture site, usually the antecubital fossa (the depression in the anterior region of the elbow (Fig. 19-3). A BP cuff (sphygmomanometer) may be used instead of a tourniquet if a patient is difficult to draw. This technique should only be performed by experienced HM.

3. Position the patient’s arm extended with little or no flexion at the elbow.

4. Locate a prominent vein by palpation (feeling). If the vein is difficult to find, it may be made more prominent by having the patient hold their arm in a downward position.

5. Cleanse the desired site with a 70% alcohol pad or povidone-iodine solution and allow it to dry.

CAUTION:
After cleaning the desired site, only the sterile needle should be allowed to touch it.

DO NOT TOUCH WITH UNSTERILE OBJECTS.

6. "Anchor" the vein by using the thumb of the free hand placing it a minimum of 1 to 2 inches below and slightly to the side of the intended venipuncture site, and pull the skin toward the wrist.

7. Using a smooth continuous motion, introduce the needle, bevel side up, into the vein at about a 15 to 30 degree angle with the skin (Fig. 19-4).
Figure 19-4.—Phlebotomy

8. Holding the vacutainer barrel with one hand, push the tube into the holder with the other hand and watch for the flow of blood into the tube.

9. The tourniquet should be removed as soon as blood flows freely into the tubes. In some difficult draw situations, the tourniquet is sometimes left on until the last tube is filled. Do not leave the tourniquet on for more than one minute.

10. Once all the specimens have been collected, remove the final tube, hold the vacutainer with one hand and release the tourniquet with the other.

11. Place a sterile gauze over the puncture site and remove the needle with a quick and smooth motion.

12. Apply pressure to the puncture site and instruct the patient to keep the arm in a straight position. Have the patient hold pressure while the tubes are labeled.

13. Take this time to invert any tubes that need to have anticoagulant mixed with the blood.

14. Specimens are to be labeled immediately after blood collection and never before. The label must be permanently attached to the tube(s) before leaving an inpatient's bedside or dismissing an outpatient.

15. Re-inspect the puncture site to make sure bleeding has stopped, and apply a bandage.

Blood tubes

It is common that one venipuncture may result in filling numerous blood tubes for more than one laboratory test. Blood tubes have standardized color-coded stopper tops to indicate which additive or anticoagulant is used in each tube. Different testing methodologies (hematology, chemistry, alcohol) are performed on different colored tubes that prepare the blood sample for testing.

It is important for every HM to understand the different tubes available at a specific command and know what each colored tube is used for. For example, a lavender-top is often used for complete blood counts (CBCs), and a red-top is often used for blood chemistry. Tubes and their uses change from one facility to the next, so it is important to understand the procedures of each facility. Refer to Table 19-1 for a list tubes and their common uses.
**BD Vacutainer® Venous Blood Collection**

**Tube Guide**

For a full line of BD Vacutainer® Specimen Collection Products, visit www.bd.com/vacutainer.

<table>
<thead>
<tr>
<th>BD Vacutainer® Tubes with BD Multicolor™ Labeled Caps</th>
<th>BD Vacutainer® Tubes with BD Conventional Stopper Caps</th>
<th>Adhesive Labels for Blood Collection*</th>
<th>Laboratory Use</th>
<th>Site Lab's Draw Volume/Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gold</td>
<td>Cut anterior and posterior sides for vein expansion.</td>
<td>5-10 mL of blood within 30 minutes.</td>
<td>5-10 mL of blood, serially cross-mixed.</td>
<td>5-10 mL of blood, serially cross-mixed.</td>
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<td>Light Green</td>
<td>Draw between 8 and 12 hours.</td>
<td>5-10 mL of blood within 60 minutes.</td>
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<td>Red</td>
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<td>5-10 mL of blood within 30 minutes.</td>
<td>5-10 mL of blood, serially cross-mixed.</td>
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<td>Orange</td>
<td>Cut anterior and posterior sides for vein expansion.</td>
<td>5-10 mL of blood within 30 minutes.</td>
<td>5-10 mL of blood, serially cross-mixed.</td>
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<tr>
<td>Gray</td>
<td>Cut anterior and posterior sides for vein expansion.</td>
<td>5-10 mL of blood within 30 minutes.</td>
<td>5-10 mL of blood, serially cross-mixed.</td>
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<tr>
<td>Black</td>
<td>Cut anterior and posterior sides for vein expansion.</td>
<td>5-10 mL of blood within 30 minutes.</td>
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<td>White</td>
<td>Cut anterior and posterior sides for vein expansion.</td>
<td>5-10 mL of blood within 30 minutes.</td>
<td>5-10 mL of blood, serially cross-mixed.</td>
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<tr>
<td>Blue</td>
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<td>Lavender</td>
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<td>Pink</td>
<td>Cut anterior and posterior sides for vein expansion.</td>
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<td>Purple</td>
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<td>Yellow</td>
<td>Cut anterior and posterior sides for vein expansion.</td>
<td>5-10 mL of blood within 30 minutes.</td>
<td>5-10 mL of blood, serially cross-mixed.</td>
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<td>Cyan</td>
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</tr>
<tr>
<td>Pink/Gray</td>
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<td>5-10 mL of blood, serially cross-mixed.</td>
<td>5-10 mL of blood, serially cross-mixed.</td>
</tr>
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<td>5-10 mL of blood, serially cross-mixed.</td>
<td>5-10 mL of blood, serially cross-mixed.</td>
</tr>
<tr>
<td>Black/Gray</td>
<td>Cut anterior and posterior sides for vein expansion.</td>
<td>5-10 mL of blood within 30 minutes.</td>
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<td>5-10 mL of blood, serially cross-mixed.</td>
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<tr>
<td>White/Gray</td>
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</tr>
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<td>Blue/Gray</td>
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<td>Green/Gray</td>
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<td>5-10 mL of blood within 30 minutes.</td>
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**Table 19.1.—Tubes and Their Common Uses**

THE MICROSCOPE

LEARNING OBJECTIVE:

Identify the parts of the microscope, and state their functions.

Before any attempt is made to view blood smears, urinary sediments, bacteria, or parasites, the HM must know the microscope. It is a precision instrument used extensively in clinical laboratories to observe objects too small to be seen by the unaided eye. Most laboratories are equipped with binocular (two-eyepiece) microscopes, but monocular (one-eyepiece) microscopes are also commonly used in the field settings. The type of microscope most often used in the laboratory is referred to as the compound microscope (Fig. 19-5).

FRAMEWORK

The framework of the compound microscope consists of four parts: arm, stage, mechanical (movable) stage, and base.

Arm is the structure that supports the magnification and focusing system. It is the handle by which the microscope is carried.

Stage is the platform on which a specimen is placed for examination. In the center of the stage is an aperture or hole that allows the passage of light from the condenser.

Mechanical (movable) Stage holds the specimen in place and is the means by which the specimen may be moved about on the stage to view the sample.

Base is the structure on which the microscope rests.

ILLUMINATION SYSTEM

Ideal illumination of a specimen viewed under the microscope requires even light distribution. The objectives must also be entirely filled with light from the condenser. To fulfill these requirements, the illumination system of the compound microscope consists of three parts: an internal light source, a condenser, and an iris diaphragm.

Internal Light Source is located in the base of the microscope providing a precise and steady source of light into the microscope.

Condenser is composed of a compact lens system and is located below the stage. It concentrates and focuses light from the light source on the specimen.

Iris Diaphragm is located on the condenser to control the amount of light and angle of light rays that will pass to the specimen and lens, which affects the overall resolution, or ability to observe and interpret a sample.

A compound microscope contains a system of lenses with sufficient magnification and resolving power (ability to show, separate, and distinguish) allowing small elements close together in a specimen to appear larger and distinctly separated. In the following sections, the compound microscope's framework, illumination system, magnification system, and focusing system will be outlined.

Figure 19-5.—Microscope
MAGNIFICATION SYSTEM

The magnification system of the compound microscope contains at least two lens systems. The two lens systems are mounted on either end of a tube called the body tube. The lens nearest the object is called the objective lens, and the lens nearest the eye is the ocular lens or eye piece.

Objective Lens is responsible for the magnification and resolution of detail in a specimen. On a compound microscope, there is usually a set of three objective lenses (or "objectives"). Each objective lens has a different focus distance and magnification power. A set of objectives normally consists of a low-power lens (approximate magnification 10X), a high-power lens (approximate magnification 40X), and an oil-immersion lens (approximate magnification 100X). The colors on each lens may vary by manufacturer; objective lenses are also marked for easy recognition: 10X, 40X, and 100X.

Revolving Nosepiece contains openings into which objective lenses are fitted, and revolves objectives into desired position.

Body Tube is a tube that permits light to travel from the objective to the ocular lens.

Ocular Lenses or eyepieces are located on top of the body tube and usually have a magnification power of 10X. To calculate the total magnification of a specimen, multiply the magnification power of the objective by the magnification power of the ocular lens. For example, the 10X ocular and the 40X objective provide a magnification of 400X.

FOCUSBING SYSTEM

Focusing is accomplished by moving the stage up or down with the coarse and fine control knobs. Whether the stage needs to be raised or lowered depends on each individual sample and objective being used. The coarse control knob is used initially to bring the specimens image into approximate focus. Once this is accomplished, the fine control knob sharpens the image.

Coarse Control Knob is the larger inner knob. Rotating the coarse control knob allows the image to appear in approximate focus.

Fine Control Knob is the smaller outer knob. Rotating this control knob renders the image clear and well-defined.

FOCUSING THE MICROSCOPE

Focusing the microscope must be done in a specific order to avoid accidentally damaging the objective lens, the specimen, or both. The process of focusing consists of adjusting the relationship between the optical system of the microscope and the object to be examined so that a clear image of the object is obtained. The distance between the upper surface of the glass slide on the microscope stage and the faces of the objective lens varies depending upon which of the three objectives is in the focusing position. It is important to obtain a focus with the low-power objective first, then change to the higher objective.

With the low-power (10X) objective in focusing position, observe the following steps in focusing.

1. The person using the microscope will be seated facing the microscope so that the ocular lenses are facing the person. The head of the person will be lowered to one side of the microscope until the eyes are approximately at the level of the stage.

2. Using the coarse adjustment knob, the body tube will be lowered until the face of the objective is within 1/4 inch of the object. Most microscopes are constructed in such a way that the low-power (10X) objective cannot be lowered and make contact with the object on the stage. While looking through the ocular, use the coarse adjustment knob to elevate the lenses until the image becomes visible. Once the object is clearly visible, the fine adjustment knob is used to obtain a clear and distinct image. The focusing knob must not be moved while changing lenses.
3. If the high-power objective (40X) is to be used next, it is brought into position by revolving the nosepiece (a distinct "click" indicates it is in proper alignment). The fine adjustment knob is used only to bring the object into exact focus.

4. If the specimen is too dark, increase lighting by opening the iris diaphragm of the condenser located at the base of the microscope on the light source.

5. The oil-immersion objective (100X) is used for detailed study of stained blood and bacterial smears. The distance between the objective lens and the object is very short. Great care must be taken to not damage the specimen. After focusing with the high-power objective and scanning for well-defined cells, turn the objective. Place a small drop of immersion oil, free of bubbles, on the slide. Center the drop in the circle of light coming through the condenser. Revolve the objective carousel to bring the oil-immersion objective into place. Do not attempt coarse adjustment at this time or the lens and specimen may be damaged. The final step in focusing is done with the fine adjustment knob. It is with this lens in particular that lighting is important. The final focus, clear and well-defined, will be obtained only when proper light adjustment is made.

CARE OF THE MICROSCOPE

The microscope is an expensive and delicate instrument that must be given proper care. Moving or transporting microscopes will be accomplished by grasping the arm of the scope in one hand and supporting the weight of the scope with the other hand under the base. Avoid sudden jolts and jars. Keep the microscope clean at all times. When not in use, microscopes should be enclosed in dustproof cover or stored in their case. Remove dust with a lint-free lens tissue.

Lenses may be wiped carefully with lens tissue. When the oil-immersion lens is not being used, remove the oil with lens tissue. Use lens cleaning solution on lenses only when required to remove dried oil and only in the minimal amount necessary. Never use alcohol or similar solvents to clean lenses since alcohol will damage the lens assembly.

COMPLETE BLOOD COUNT

LEARNING OBJECTIVES:

Identify the parts of a complete blood count.

Identify the normal values for each part.

A complete blood count routinely consists of the following tests:

- Total red blood cell (RBC) count
- Hemoglobin determination (Hgb)
- Hematocrit calculation (Hct)
- Total white blood cell (WBC) count
- White Blood Cell Differential count

The complete blood count, commonly referred to as a CBC, is used in the diagnosis of many diseases. Blood collected for these tests are capillary or venous blood. CBCs may be performed either manually or by using automated hematology analyzers. The manual methods performed by laboratory technicians and Independent Duty Corpsmen are used in isolated locations and onboard some Naval vessels where a hematology analyzer is not practical.

RED BLOOD CELL COUNTS

The red cell count is used in the diagnosis of many diseases. A red cell count that drops below normal values may indicate anemia. A red cell count that rises above the normal values may indicate dehydration.
Manually counting red blood cells (erythrocytes) is a time-consuming procedure that provides only limited value in an operational setting, so it is no longer included in this manual.

HEMOGLOBIN DETERMINATION

A routine test performed on practically every patient is the Hemoglobin determination, or hemoglobinometry. It is the measurement of the concentration of hemoglobin within the patient’s red blood cells. The primary function of hemoglobin is delivery and release of oxygen to the tissues and facilitation of carbon dioxide excretion. The formation of hemoglobin takes place during the development of red cells located in bone marrow.

Values are affected by age, sex, disease, and altitude. Different situations affect the function of hemoglobin in different ways. For example, iron deficiency anemia may drop hemoglobin from a normal value to a critically low value. Above-normal values may occur when dehydration develops. Changes in altitude affect the oxygen content of the air and, therefore, also affect hemoglobin values. At higher altitudes there is less oxygen in the air, resulting in an increase in red cell counts and hemoglobin values. At lower altitudes there is more oxygen, resulting in a decrease in red cell counts and hemoglobin values.

The normal values for hemoglobin determinations are:

Grams per 100 ml blood

Woman ...... 12 to 16
Men ...... 14 to 18

In manual methods for determining blood hemoglobin, blood is mixed with cyanmethemoglobin. This process hemolyzes, or destroys the red cells, disrupting the integrity of the red cells' membrane and causing the release of hemoglobin, which, in turn, is converted to a brownish-colored solution which is then compared with a color standard.

HEMATOCRIT (PACKED CELL VOLUME) DETERMINATION

The hematocrit, or packed RBC volume, is the ratio of the volume of RBCs to the volume of whole blood. It is usually expressed as a percentage. The normal values for hematocrit determinations are:

Percentage of blood volume

Woman .......... 37 to 47%
Men .......... 42 to 52%

When hematocrit determinations are below normal, medical conditions such as anemia may be present. Above-normal hematocrit determinations indicate medical conditions like dehydration.

Automated hematology analyzers currently supply most hematocrits. When hematology analyzers are not available, determinations can be manually performed by the microhematocrit method. This method calls for the blood to be centrifuged. The percentage of packed red cells is found by calculation and reported as a percentage.

TOTAL WHITE BLOOD CELL COUNT

The total white cell (leukocyte) count determines the number of white cells per cubic millimeter of blood. A great deal of information can be derived from white cell studies. The white blood cell (WBC) count and the differential count are common laboratory tests, and they are almost a necessity in determining the nature and severity of systemic infections. Normal WBC values in adults range from 4,800 to 10,800 cells per cubic millimeter; reported as 4.8 to 10.8 X 10⁹/ml (per cubic millimeter).

White blood cell counts are performed either manually or with automated hematology analyzers. Only the manual method will be covered in this chapter. After a brief introduction on abnormal white blood cell counts, the Unopette method will be covered for manually counting white blood cells.
Abnormal White Cell Counts

When the WBC rises above normal values, the condition is referred to as leukocytosis. Leukocytosis frequently occurs when systemic or local infections (usually due to bacteria) are present. In severe medical conditions, white cell counts will exceed 50,000/mm³.

Counts for infections are highly variable based on each individual patient, onset of infection, and a patient’s individual response. Other physiological conditions that can cause leukocytosis may occur as follows:

- Shortly after birth
- Pregnancy
- Appendicitis
- Ulcers
- Emotional stress
- Anxiety
- Strenuous exercise

An abnormally low white cell count, known as leukopenia, may be caused by the following conditions:

- Severe or advanced bacterial infections such as typhoid, paratyphoid, and sometimes tularemia
- When a bacterial infection has been undetected for a period of time
- Infections caused by viruses and rickettsiae, such as measles, rubella, smallpox, infectious hepatitis, psittacosis, dengue fever, and influenza
- Protozoal infections (such as malaria) and helminthic infections (such as trichinosis)
- Overwhelming infections when the body’s defense mechanisms break down
- Anaphylactic shock
- Radiation

WHITE BLOOD CELL DIFFERENTIAL COUNT

A total WBC is not necessarily indicative of the severity of a disease, since some serious ailments may show a low or normal overall white cell count. For this reason, a differential white cell count is performed. This consists of an examination of blood to determine the presence and the number of different types of white blood cells usually expressed in a percentage. This study often provides more helpful information in determining the severity and type of an infection than any other single procedure used in the examination of the blood.

The role of white blood cell is to control various disease conditions. Although these cells do most of their work outside the circulatory system, they use the blood for transportation to sites of infection. The amount and type of cells in the circulating blood can provide valuable information about the body’s immediate response to infection or disease.

Five types of white cells are normally found in the circulating blood. They are:

- Neutrophils
- Eosinophils
- Basophils
- Lymphocytes
- Monocytes

Cell Identification

To perform a differential white cell count, a laboratory technician or HM must be able to identify the different types of white blood cells. The ability to properly identify the different types of white cells is not difficult to develop, but does require a thorough knowledge of staining characteristics and morphology (the study of the form and structure of organisms). This knowledge can be gained only by extensive, supervised practice, but an introduction is included in this chapter for a better understanding.
Laboratories use a blood smear to obtain a differential white cell count. To prepare a blood smear, a blood specimen is spread across a glass slide, stained to enhance leukocyte identification, and examined microscopically. Material requirements and the step-by-step procedure for performing a blood smear will be covered later in this chapter.

**NEUTROPHILS.**—account for the largest percentage of leukocytes found in a normal blood sample, and function by ingesting invading bacteria. On a stained blood smear, the cytoplasm of a neutrophil has numerous fine, barely visible lilac-colored granules and a dark purple or reddish purple nucleus (Fig. 19-6). The nucleus may be oval, horseshoe or “S”-shaped, or segmented (lobulated). They are subclassified according to their age or maturity, which is indicated by changes in the nucleus.

**Neutrophilic Band.**—sometimes called a "stab" cell, is an older or intermediate neutrophil. It has started to elongate and has curved itself into a horseshoe, C or U-shape. As the band ages, it matures into a segmented neutrophil (Fig. 19-7).

![Figure 19-7.—Neutrophilic Band](image)


**Segmented Neutrophil.**—is a mature neutrophil. The nucleus of a segmented neutrophil is separated into two, three, four, or five segments or lobes.

![Figure 19-6.—Neutrophil](image)

EOSINOPHIL.—The function is to destroy parasites and respond in immediate allergic reactions. The cytoplasm of an eosinophil contains numerous large reddish-orange granules (Fig. 19-8). The most common cause of increased eosinophils worldwide is parasitic, in particular helminthic, infections.

![Eosinophil Image](image)

**Figure 19-3.—Eosinophil**

Image reprinted with permission from:

LYMPHOCYTE.—The function is associated with immune response and the body's defense against viral infection. The cytoplasm of a lymphocyte is clear sky blue, scanty, with few unevenly distributed, blue granules with a halo around them (Fig. 19-10). It is generally round, oval, or slightly indented, and the chromatin (a network of fibers within the nucleus) is lumpy and condensed at the periphery.

![Lymphocyte Image](image)

**Figure 19-10.—Lymphocyte**

Image reprinted with permission from:

BASOPHIL.—A rise in basophils is associated with inflammatory disorders and certain leukemias. Scattered deep bluish-purple granules that are darker than the nucleus, characterize the cell as a basophil (Fig. 19-9). Granules may overlay the nucleus as well as the cytoplasm.

![Basophil Image](image)

**Figure 15-9.—Basophil**

Image reprinted with permission from:
MONOCYTE.—The largest of the normal white blood cells, controls microbial and fungal infections, and removes damaged cells from the body. The monocyte has an indented nucleus and an abundant pale bluish-gray cytoplasm (Figs. 19-11 and 19-12).

BACTERIOLOGY

LEARNING OBJECTIVE:

Identify bacteria classifications, common bacteria, and procedural steps for making smears, performing Gram staining, and reading and reporting smears.

Bacteriology is the study of bacteria. Of primary interest to the HM is medical bacteriology, which deals with the bacteria that cause disease in man. Bacteria are found almost everywhere, and the human body harbors vast numbers. Many bacteria are beneficial and essential to human life; only a few are harmful to man.

BACTERIA CLASSIFICATION

Since there are thousands of types of bacteria, a method of classification is essential. Bacteria are classified according to their respective:

- Disease-producing ability
- Growth requirements
- Morphologic characteristics
- Toxins produced
- Gram’s stain reaction

Disease-Producing Ability

The disease-producing ability of bacteria is referred to as either pathogenic or nonpathogenic. Pathogens are bacteria that cause diseases, and nonpathogens are harmless bacteria. Bacteria that are essential to the body are, in their proper environment, called common or normal flora. For example, certain bacteria in the throat are normal flora, but when found elsewhere (such as in the blood stream, possibly as a result of tooth extraction), they may cause diseases such as septicemia and endocarditis.
Growth Requirements

The four growth requirements for bacteria are:

- Temperature
- Oxygen
- Nutrition
- Moisture

TEMPERATURE REQUIREMENTS.— Temperature requirements are divided into the following three categories:

- Psychrophilic "cold loving" bacteria that reproduce best at low temperatures (4°C)
- Mesophilic bacteria that reproduce best at body temperature (35 °C) and are the primary pathogens in man
- Thermophilic bacteria that reproduce best at higher temperatures (42 °C)

OXYGEN REQUIREMENTS.— The amount of oxygen needed for an organism to grow or reproduce varies with the type of organism. Aerobes are organisms that reproduce in the presence of oxygen. Anaerobes are organisms that do not reproduce in the presence of oxygen. Other bacteria have varying oxygen requirements.

NUTRITION REQUIREMENTS.— Nutrition requirements for the various types of bacteria depends on what particular environment is required in the body or laboratory setting.

MOISTURE REQUIREMENTS.— Moisture is indispensable for bacterial growth providing an environment for metabolic reactions to take place.

Morphologic Characteristics

The structural (or morphologic) characteristics of bacteria are based on three distinct shapes or categories:

- Coccus (pl. cocci) spherical, appears singly, in pairs, chains, clusters, or packets
- Bacillus (pl. bacilli) rod-shaped, appears singly, in chains, or in different organizations, (i.e. railroad tracks or school of fish)
- Spirochetes (pl. spirilla) helical, spiral, corkscrew-shaped, appearing singly only

Toxins Produced

Generally, toxins produced are waste products of metabolism in a bacterial cell. Some bacteria produce toxins that cause febrile or fatal reactions. Toxins are divided into two categories:

- Endotoxin - Comprises part of the cell wall and is released as the bacterial cell is destroyed. Endotoxins are less potent than exotoxins, but may affect the patient during the course of antibiotic therapy
- Exotoxin - Produced by bacteria and found outside the bacterial cell in the surrounding medium. Exotoxins are highly poisonous and associated with septic shock

Gram Stain Reaction

A gram-stained smear is a key diagnostic tool. It can detect bacteria in patient specimens and is critical in identifying cultured bacteria. This differential stain is based on the cell wall differences between gram positive and gram negative bacteria. Gram positive cells retain the primary crystal violet stain during decolorization and retain the violet stain; gram negative do not, and are counterstained pink. The basic staining and morphological characteristics (gram negative rods, gram positive cocci, gram negative diplococci) provide an initial classification of bacteria that enable selection of the correct antibiotic therapy.
COMMON BACTERIA

Bacteria are named by genus and species. The first word (capitalized) indicates the genus; the second word (not capitalized) indicates the species, a subdivision of the genus. For example:

**GENUS SPECIES**
*Neisseria gonorrhoeae*

Table 19-2 provides familiarization with commonly encountered bacteria. This table lists the bacteria's morphologic shape, Gram stain response, genus and species, and the type of infection it produces.

<table>
<thead>
<tr>
<th>Morphologic Shape</th>
<th>Gram-Positive or -Negative</th>
<th>Genus &amp; Species</th>
<th>Type of Infection</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cocci</td>
<td>Positive</td>
<td><em>Streptococcus pneumoniae</em></td>
<td>Pneumonia</td>
</tr>
<tr>
<td></td>
<td></td>
<td><em>Streptococcus pyogenes (Beta</em>&lt;br&gt;Streptococci Group A)*</td>
<td>Strep throat</td>
</tr>
<tr>
<td></td>
<td></td>
<td><em>Staphylococcus aureus</em></td>
<td>Boils, furuncles, osteomyelitis, pneumonia, septicemia, endocarditis, and impetigo</td>
</tr>
<tr>
<td>Negative</td>
<td>Neisseria gonorrhoeae</td>
<td></td>
<td>Gonorrhea</td>
</tr>
<tr>
<td></td>
<td>Neisseria meningitidis</td>
<td></td>
<td>Meningitis</td>
</tr>
<tr>
<td>Bacilli</td>
<td>Positive</td>
<td><em>Corynebacterium diphtheriae</em></td>
<td>Diphtheria</td>
</tr>
<tr>
<td></td>
<td></td>
<td><em>Clostridium (all are anaerobic and spore producers)</em>&lt;br&gt;perfringens (welchii)&lt;br&gt;tetani&lt;br&gt;botulinum</td>
<td>Gas gangrene&lt;br&gt;Tetanus&lt;br&gt;Botulism</td>
</tr>
<tr>
<td>Negative</td>
<td>Yersinia (Pasteurella) pestis</td>
<td></td>
<td>Bubonic plague</td>
</tr>
<tr>
<td></td>
<td>Brucella abortus</td>
<td></td>
<td>Brucellosis</td>
</tr>
<tr>
<td></td>
<td>Bordetella pertussis</td>
<td></td>
<td>Whooping cough</td>
</tr>
</tbody>
</table>

Table 19-2.—Common Pathogenic Bacteria

19-20
Smear

Smears may be prepared from clinical specimens (i.e. discharge or CSF) or from culture media spread across a glass slide for microscopic examination. To enhance the visualization of microorganisms on the smear, Gram staining is used. Once the smear is stained, it is ready to be examined under the microscope. Normally, smears are examined by laboratory technicians who report their findings.

Gram-positive organisms are easy to see because they stain a deep blue or blue-black (Fig. 19-13).

Gram-negative organisms stain a deep pink, but since the background material is also pink, minute and detailed inspection is necessary before reporting the results (Fig. 19-14).

![Figure 19-14.—Gram Negative Organisms](image)

In the presence of gonorrhea (caused by Neisseria gonorrhoeae), the smear will reveal varying intracellular and extracellular gram-negative, bean-shaped cocci in pairs (diplococci). Such a finding could be considered diagnostic for gonorrhea in urethral discharge from symptomatic males. It is important to point out that only a few of the many WBCs on the slide may contain bacteria, and sometimes it requires considerable search to find one.
SEROLOGY

LEARNING OBJECTIVE:

Identify principles and procedures for the Rapid Plasma Reagin (RPR) Card Test and the Monospot Test.

Serology consists of procedures by which antigens and reacting serum globulin antibodies may be measured qualitatively and quantitatively. These tests have been devised to detect either antigens present or antibodies produced in a number of conditions. Most tests are based on agglutination reactions between an antigen and a specific antibody. The reactions result in a visual clumping of test solution when antigens and antibodies react. The HIM may have the opportunity to perform simple screening serology tests.

An antigen is a substance that, when introduced into an individual's body is recognized as foreign by an individual's immune system and causes a detectable reaction.

Antibodies are specific defensive proteins produced when an antigen stimulates individual cells. The primary function of an antibody in body defenses is to combine with antigens. Antibodies are produced by the host in response to the presence of an antigen and are capable of reacting with antigens in some detectable way.

The antigen-antibody reaction takes place when antibodies bind with specific antigens depending on a close three-dimensional fit. Agglutination tests are widely used to detect and measure the presence of antigen-antibody reactions.

Principles and procedures of two serologic tests, the Rapid Plasma Reagin (RPR) card test and the Monosticon DRI-DOT® Slide Test are covered in the following sections.

RAPID PLASMA REAGIN (RPR) CARD TEST

The RPR Card test is a non-specific, easily performed screening test for syphilis. Reactions are occasionally observed with other acute and chronic conditions associated with tissue damage. Everything needed for the test is in a kit that is available commercially. This test kit is very useful aboard ship and at small Naval station settings. The kit is standard throughout Navy medicine.

Principle of the RPR Card Test

In the RPR Card test method of syphilis detection, a specific antigen (carbon-particle cardiolipin) detects "reagin," a substance present in the serum of persons who are infected with syphilis or suffering from similar tissue damage. Reagin is usually developed 1-4 weeks after the appearance of a primary chancre. Reactive specimens appear as black clumps against a white background. Nonreactive specimens appear as an even, light-gray color.

The RPR is a non-specific screening test and is not reported as positive or negative for disease. It is reported as reactive or non-reactive.

MONOSPOT TEST

Mononucleosis imitates many diseases so well that diagnosis is confirmed only by selective serologic testing. The Monospot Test is an accurate, 2-minute disposable test designed to detect the presence of infectious mononucleosis antibodies in serum, plasma, or whole blood. Although there are numerous tests for infectious mononucleosis, all have the same principle, and similar methods.

Principle of the Monospot

The Monospot Test consists of specially prepared, stable sheep or horse erythrocyte antigen (dyed) and guinea pig antigen on a disposable slide. When serum, plasma, or whole blood is mixed with these antigens on the slide, the test result for infectious mononucleosis will be positive or negative.
A positive result is indicated by agglutination, or clumping, and a negative result is indicated by no agglutination (Fig. 19-15). A negative monospot may not necessarily rule out the presence of infectious mononucleosis.

![Positive Agglutination vs Negative Agglutination](image.png)

**Figure 19-15.—Illustration of Positive and Negative Monostotic DRI DOT® Slide Test Results**

**FUNGUS TEST**

**LEARNING OBJECTIVE:**

*Identify how potassium hydroxide (KOH) preparation is used in the detection of fungi.*

Fungi (sing. fungus) are chlorophyll-free, heterotrophic (not self-sustaining) of the same family of plants as algae and lichens. They reproduce by spores that germinate into long filaments called hyphae. As the hyphae continue to grow and branch, they develop into a mat of growth called the mycelium (pl. mycelia). From the mycelium, spores are produced in characteristic patterns. These spores, when dispersed to new substances, germinate and form new growths. Reproduction is often asexual, usually by budding (as in yeast), but certain fungi have sexual reproduction. Common superficial infections of the skin caused by fungi are athlete’s foot and ringworm of the scalp.

A simple and frequently used method of detecting fungi is the potassium hydroxide (KOH) preparation. Fungi are seen in clustered round buds with thick walls, accompanied by fragments of mycelia.

Scrapings from the affected area of the skin are mounted in commercially prepared 10% KOH for positive laboratory diagnosis.

To detect fungi in infected tissue using the KOH preparation, follow the steps below:

1. Place skin, hair, or nail scrapings from the affected area on a glass slide and add one drop of 10% KOH.
2. Place a coverslip on the preparation.
3. Warm the preparation gently over the tip of a flame, being careful not to boil it, and allow it to stand until clear. Do not allow the preparation to dry out.
4. Examine the preparation by using the high-power objective on microscope with subdued light.
   a. Fungi on the skin and nails appear as refractile, or reflective, fragments of fungal elements.
   b. Fungi in the hair appear as dense clouds around the hair stub or as linear rows inside the hair shaft.

**URINALYSIS**

**LEARNING OBJECTIVES:**

*Identify the three types of urine specimens.*

*Identify the methods used to preserve urine specimens.*

*Identify the steps for performing a urinalysis.*

The analysis of urine is considered the beginning of laboratory medicine. Urine is readily available, easily collected, and contains information about many of the body’s major metabolic functions.

The physical and chemical properties of normal urine are constant and abnormalities are easily detected. The use of simple tests provides the provider with information for the diagnosis and management of many diseases.
This section outlines the three types of urine specimens, methods used to preserve urine specimens, the procedure for performing a routine and microscopic examination of urine specimens, and some simple interpretations of the findings.

**URINE SPECIMENS**

Urine specimens for routine examinations must be collected in aseptically clean containers. Unless circumstances warrant, avoid catheterization as it may cause a urinary tract infection. Specimens of female patients are likely to be contaminated with albumin (protein) and blood from menstrual discharge, or with albumin and pus from vaginal discharge. For bacteriologic studies, care must be taken to ensure that the external genitalia have been thoroughly cleansed as directed by laboratory procedures for clean-catch specimen collection. The patient must void the initial stream of urine into the toilet or a suitable container and the remainder directly into a sterile container. All urine specimens should either be examined when freshly voided, or refrigerated to prevent decomposition of urinary constituents and to limit bacterial growth. The following sections will cover three types of urine specimens: random, first morning, and 24-hour.

**Random Urine Specimen** is the most commonly received specimen because of the ease of collection and convenience for the patient. These specimens are collected without regard to the time of day or fasting state. This sample is useful for routine screening tests to detect obvious abnormalities. It may produce erroneous results caused by dietary intake or physical activity just prior to the collection of the specimen. It is the least valid specimen, and patients may later be requested to collect additional specimens under more controlled conditions.

**First Morning Urine Specimen** is the first urine voided upon rising. It is the ideal screening specimen, because it is usually concentrated and more likely to reveal abnormalities. If positive results are obtained from the first morning specimen, the physician may order a 24-hour specimen for quantitative studies.

**Twenty-Four Hour Urine Specimen** measures the exact output of urine over a 24-hour period. Use the following steps to collect this specimen.

1. Have patient empty bladder early in the morning and record time. Discard this urine.
2. Collect all urine voided during next 24 hours.
3. Instruct patient to empty bladder at 0800 the following day (end of 24-hour period). Add this urine to pooled specimen.

Refrigerate specimen during collection. Depending on the test being performed, add a preservative to the first specimen voided.

The normal daily urine volume for adults ranges from 600 to 2,000 ml, averaging about 1,500 ml. The amount of urine excreted in 24 hours varies with fluid intake and the amount of water lost through perspiration, respiration, and bowel activity. Diarrhea or profuse sweating reduces urinary output; diabetes is associated with increased urinary output.
PRESERVATION OF URINE SPECIMENS

To delay decomposition of urine, use the following methods of preservation:
- Refrigeration
- Preservatives
  - Hydrochloric acid
  - Other preservatives as directed by laboratory staff

NOTE:
Before adding a preservative to a urine specimen, contact the laboratory performing the testing to find out what preservative and quantity to use. Preservative requirements vary from laboratory to laboratory.

ROUTINE URINE EXAMINATION

A routine urinalysis includes the examination of physical characteristics, chemical characteristics, and microscopic structures in the sediment. A sample for urinalysis (routine and microscopic) should be at least 12 ml in volume (adult), and either a random or first morning specimen. Children may only be able to provide a small volume, but 10-15 ml is preferred.

Physical Characteristics

Physical characteristics evaluated during a routine urinalysis include color, appearance, and specific gravity.

COLOR - The normal color of urine varies from straw to amber. Diluted urine is generally pale; concentrated urine tends to be darker. The following are terms used to describe the color of urine:
- Colorless
- Light straw
- Straw
- Dark straw
- Light amber
- Amber
- Red, abnormal color

The color of urine may be changed by the presence of blood, drugs, or diagnostic dyes. Examples are:
- Red or red-brown - caused by the presence of blood
- Yellow or brown (turning greenish with yellow foam when shaken) - caused by the presence of bile
- Olive green to brown-black - caused by phenols (an extremely poisonous compound, used as an antimicrobial agent)
- Dark orange - caused by Pyridium® (a topical analgesic used in the treatment of urinary tract infections)

CLARITY

Urine's appearance may be reported as clear, hazy, slightly cloudy, cloudy, or turbid. Freshly passed urine is usually clear or transparent. Urine can appear cloudy when substances such as blood, leukocytes, crystals, pus, or bacteria are present. A report of transparency is of value only if the specimen is fresh. After standing, all urine becomes cloudy because of decomposition, salts, and the action of bacteria. Upon standing and cooling, all urine specimens will develop a faint cloud composed of mucus, leukocytes, and epithelial cells. This cloud settles to the bottom of the specimen container and is of no significance.

SPECIFIC GRAVITY

The ability of the kidneys to selectively reabsorb essential chemicals and water is one of the body's most important functions. Specific gravity is defined as the density of a solution compared to an equal volume of distilled water. The specific gravity varies directly with the amount of solids dissolved in the urine, and normally ranges from 1.015 to 1.030 during a 24-hour period.
The first morning specimen of urine is more concentrated and will have a higher specific gravity than a specimen passed during the day. A high fluid intake may reduce the specific gravity to below 1.010. In the presence of disease, the specific gravity of a 24-hour specimen may vary from 1.001 to 1.060.

Specific gravity is measured with an index refractometer, available as standard equipment at most duty stations (Fig. 19-16). It may be held manually or mounted on a stand like a microscope. The specific gravity of urine is determined by the index of light refraction through solid material.

**Chemical Characteristics**

Chemical characteristics evaluated during a routine urinalysis include pH, protein, glucose, ketones, blood, bilirubin, urobilinogen, nitrite, leukocytes, and specific gravity depending on the test strip used. Each manufacturer of reagent strips includes a color chart for multiple chemical determinations. The strip is dipped into the urine specimen and compared to the color values for the various tests on the accompanying chart. The color chart also indicates numerical pH values, which should be reported. Since this test is based on color changes, those with color deficiency may need to take extra care to interpret the results.

**Urinalysis Reagent Strips**

Reagent strips consist of chemically impregnated absorbent pads attached to a plastic strip. A color-producing chemical reaction takes place when the absorbent pad comes in contact with urine. Color reactions are interpreted by comparing the color produced on the pad with a chart supplied by the manufacturer of the reagent strips. Several colors or intensities of a color for each substance being tested appear on the chart. By careful comparison of the colors on the chart and the strip, a semi-quantitative value of trace, 1+, 2+, 3+, or 4+ can be reported.

Testing methodology includes dipping the reagent strip completely, but briefly, into a well-mixed specimen; removing the excess urine from the strip when withdrawing it from the specimen; waiting the specified length of time for reactions to take place; and comparing the colored reactions against the manufacturer's chart using a good light source.

Improper technique can result in errors. Formed elements such as red and white blood cells sink to the bottom of the specimen and will be undetected in an unmixed specimen. Allowing the strip to remain in the urine for an extended period may cause leaching of reagents from the pads. Likewise, excess urine remaining on the strip after its removal from the specimen can produce a run-over between chemicals on adjacent pads, producing distortion of the colors.
To ensure against run-over, blotting the edge of the strip and holding the strip horizontally while comparing it with the color chart is recommended. The amount of time needed for reactions to take place varies from an immediate reaction to two minutes. The required reaction time is written on the comparison chart located on the side of the reagent strip container (Fig. 19-17).

All bottles are stamped with an expiration date that represents the functional life expectancy of the chemical pads. Reagent strips should not be used past the expiration date. Care must be taken not to touch the chemical pads when removing the strips.

**Microscopic Examination of Urine Sediment**

Microscopic examination of urine sediment at 40X is usually performed in addition to routine chemical procedures. This examination requires a degree of skill acquired through practice under the immediate supervision of an experienced technician. The specimen used for microscopic examination should be as fresh as possible. Red cells and many formed solids tend to disintegrate upon standing, particularly if the specimen is warm or alkaline.

The strip must be held close to the color chart without actually placing it on the chart. Reagent strips and color charts from different manufacturers are not interchangeable. Specimens that have been refrigerated must be allowed to return to room temperature prior to reagent strip testing, since some enzymatic reactions on the strips are temperature dependent.

In addition to the use of correct testing technique, reagent strips must be protected from deterioration caused by moisture, volatile chemicals, heat, and light. Reagent strips are packaged in opaque containers with a desiccant to protect them from light and moisture. Strips are removed just prior to testing, and the bottle is tightly resealed immediately after strips are removed.

*Figure 19-17.—Urine Dipstick*

*Used with permission of Siemens Healthcare Diagnostics Inc.*
CLINICALLY SIGNIFICANT FINDINGS—
Leukocytes, erythrocytes, and casts may all be of clinical significance when found in urine sediment.

Leukocytes.—Normally, 0 to 3 leukocytes per high-power field will be seen on microscopic examination (Fig. 19-18). More than 3 cells per high-power field may indicate disease somewhere in the urinary tract. Estimate the number of leukocytes present per high-power field and report it as the "estimated number per high-power field."

Epithelial Cells.—It is not unusual to find epithelial cells (Fig. 19-20) in the urine as they are derived from the linings of the genitourinary system. Unless they are present in large numbers or in abnormal form, they represent the normal sloughing of old cells (except that the nuclei become more distinct).
**Casts.**—These urinary sediments are formed by coagulation of albuminous material in the kidney tubules. Casts are cylindrical and vary in diameter (Figs. 19-21 and 19-22). The sides are parallel, and the ends are usually rounded. Casts in the urine always indicate some form of kidney disorder and should always be reported. If casts are present in large numbers, the urine is almost sure to be positive for albumin.

**RESULTS AND READINESS**

**LEARNING OBJECTIVES:**

*Explain the clinical significance of critical results.*

*Identify the significance of laboratory function as they relate to operational readiness.*

**CRITICAL RESULTS**

Some test results that the HM may encounter in performing routine testing are indicative of life-threatening conditions that need to be communicated to the provider immediately. Each treatment facility has specific guidance for critical results. Below are results that will always be considered critical regardless of location.

- White Blood Cell (WBC) count above 50,000 indicates acute infection
- Hemoglobin concentration below 7 indicates severe anemia that may require a transfusion
- Glucose and ketones both positive on the urine reagent strip may indicate uncontrolled diabetes
- Bacteria present in a gram stain from direct patient smears

If the HM encounters any of the above results, they should report the results to a provider immediately. Other critical test results may be defined locally and should be included in the laboratory department Standard Operating Procedures manual along with reporting procedures.
Walking Blood Bank\textsuperscript{22} (WBB)

Whole blood transfusions may be required in an emergency situation. The Walking Blood Bank (WBB) will be used in a mass casualty situation if necessary and feasible in the operational setting. WBB donors should only be used in a true emergency when the delay necessary to transfer the patient to a shore-based medical facility would be detrimental to a critical patient. The WBB is established on numerous operational platforms by maintaining a list of all personnel eligible as blood donors (walking blood bank). A minimum file of ten percent of certain ship's company is required to be enrolled in the walking blood bank.

Although transfusion at sea is a rare event, the availability of a well-planned transfusion program is required, and will be coordinated by the Senior Medical Department Representative and embarked laboratory personnel. The life-saving nature of blood products requires strict regulations and oversight. HM's may be requested to volunteer as members of the WBB, recruit members of the operational platform for the WBB, or assist the laboratory staff when directed by the surgeon to initiate the WBB in an emergency or mass casualty situation.

LABORATORY RESULTS AND READINESS\textsuperscript{22}

Certain laboratory results are tracked for individual, medical, and operational readiness.

G6PD, Sickle Cell, ABO/Rh (Blood Type), and DNA Reference specimen are usually collected and documented upon creation of treatment record at point of accession.

Current HIV- Requirement varies based on platform.

It is important for treatment records to be adequately screened upon a Sailor's reporting to each new command to document these items into the appropriate readiness tracking programs. If deficiencies are found, these tests should be recollected, tested, and documented as soon as possible.