 FIRELANDSHEALTH <small>Irelands Regional Medical Center</small>	Title: COLLECTION OF SPECIMENS FOR MICROBIOLOGICAL EXAMINATION	ID #: LAB.403.400 Effective: 01/86 Page: 1 of 14
	STANDARD POLICY AND PROCEDURE FORM	

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Microbiology Specimen Collection (Quick Reference Guide)

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A. GENERAL REQUIREMENTS

The success of the bacteriologic procedures depends to a great extent on the manner on which the specimens are obtained and the promptness with which they are taken to the Laboratory. Such procedures are more often than not made valueless by lack of care and faulty methods used in the collection and handling of specimens. The following are some general rules that should be followed.

1. All specimens are to be labeled with the patient's name, date of birth, source of specimen, and the time and date of collection. On the requisition, it should be stated if the patient is on antibiotic therapy and what type he/she is on. The specimen should be sent to the lab immediately.
2. Swab specimens should be sent using the Starswab II transport system. Dry swabs are not acceptable for culture. Specimens on the Starswab are stable for 24 hours.
3. Anaerobes are to be transported in such a way as to prevent exposure of the specimen to oxygen. This can be done by use of the Starswab II transport system. Anaerobic transport media is available for tissue specimens in the Microbiology department.
4. Specimens should be obtained, if at all possible, before antibiotics or antimicrobial agents are administered. Specimens must be collected with a minimum of contamination from adjacent tissues or secretions.
5. A sufficient quantity of specimen must be obtained to perform the culture techniques requested. A collection of 24-hour urine should not be sent to the Laboratory for routine culturing, as it will be overgrown with normal microorganisms that grow on the skin.
6. All specimens are to be submitted to the Laboratory within 2 hours of collection. Please see the Microbiology procedure "Specimen Rejection" for further instructions.
7. If a sample is submitted on a Starswab II Culture Collection and Transport System, and plating is delayed, the storage (room temperature or refrigeration) should not exceed 24 hours. It has been found that refrigeration (2-8 C) is the preferred storage if storage is needed beyond the 24 hour period.
8. Because urine is an excellent culture media, urines should be cultured as soon as possible after collection. Do not allow urines to remain at room temperature for more than two hours between collection and culturing. If urines cannot be cultured upon receipt in the laboratory, they can be refrigerated for up to 24 hours.
9. All samples should be in a tightly sealed container with no external spillage.
10. Specimens submitted for routine culture, fungus culture and/or acid fast culture need a separate specimen submitted for each culture type.

B. COLLECTION PROCEDURES

The following are general collection procedures for various specimens. These are the best ways recommended for their collection to give accurate results.

Please Note: **Gloves should be worn during collection of all Microbiology specimens.**

1. Throat Cultures

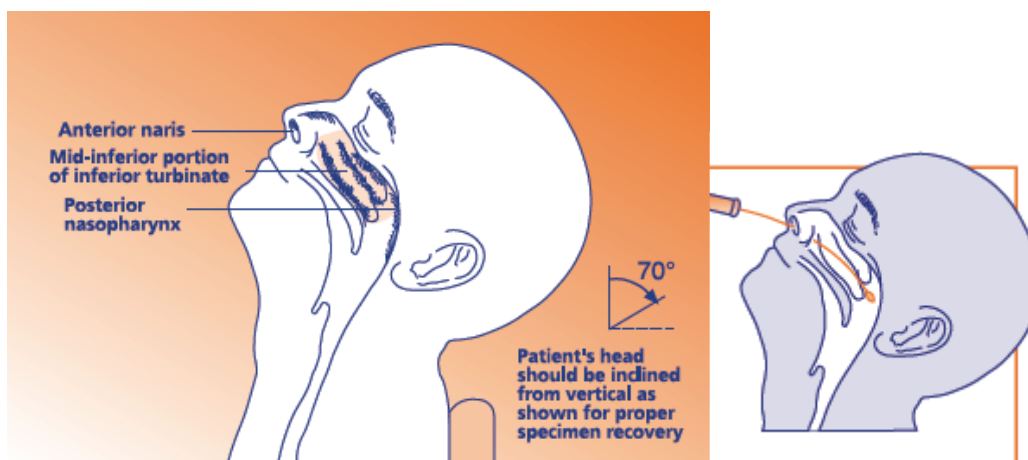
In order to avoid contamination with the saliva and normal flora of the mucous membranes, the swab should be carefully inserted through the mouth with the aid of a strong light, with the tongue depressed. Rub the swab over each tonsillar area and the posterior pharynx. Any area with exudate should also be touched. Do not contaminate the swab by touching the tongue and lips. If the doctor is looking for specific organisms, such as Neisseria gonorrhoeae, this should be noted on the requisition.

2. Nasal Cultures

(May be collected by doctors, nurses, and trained hospital personnel.)

Nasopharyngeal Collection:

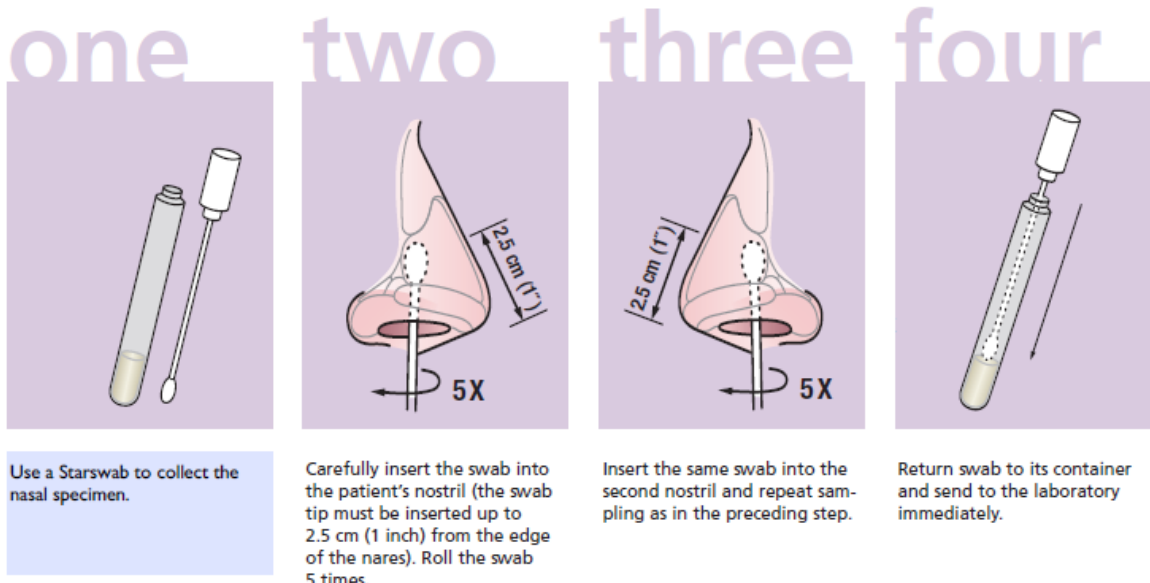
1. Insert flocked nasopharyngeal swab into one nostril until you reach resistance. Do not push any further as you may cause damage. Repeating for the second nostril will deliver optimal combined sample.
2. Rotate swab over surface of posterior nasopharynx.
3. Withdraw swab from collection site and insert into transport tube containing 1.0 ml Normal Saline or Viral Transport Media.
4. Transport to the lab as rapidly as possible.



Nasal Collection for MRSA Screening:

MRSA SCREENING

Collection Guidelines



3. Sputum Cultures


For culture, sputum should be collected in disposable plastic containers. The major problem confronting the diagnostic laboratory in regard to sputum culturing is the neglect and carelessness with which sputum collection is treated. As a result of improper collection, the number of sputum specimens submitted for culture that are primarily saliva is high. Purulent or mucopurulent material is needed for sputum culture and it should be obtained on awakening if possible. Deep coughing followed by expectoration will result in adequate material. Please note that a delay in processing of more than 2 hours may result in loss of recovery of fastidious pathogens, such as *S. pneumoniae*, and overgrowth of oronasal microbiota. Specimens should be maintained at room temperature and transported to the laboratory within 2 hours.

4. Urine Cultures

Because urine is an excellent culture media, urines should be cultured as soon as possible after collection. Do not allow urines to remain at room temperature for more than two hours between collection and culturing. If urines cannot be cultured upon receipt in the laboratory they can be refrigerated for up to 24 hours.

BD gray top tubes that state "urine C&S preservative" are acceptable for urine cultures.

*****SEE NEXT PAGE*****

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A. Clean voided midstream samples

Clean Catch / Midstream Urine Collection Instructions for Patients (ALSO FOUND AT END OF THIS PROCEDURE)

1. Female

For proper collection of a clean voided midstream sample from female patients, the area around the vagina and urethra should be cleansed thoroughly before specimen collection. The area should be wiped from front to back to avoid spreading bacteria from the anal area.

The skin folds (labia) should be held apart during this collection.

Collect a midstream sample by urination into a toilet bowl then, without stopping the stream, inserting the collection cup to collect the midstream sample. Fill the container about half full.

2. Male


For proper collection of clean voided samples from male patients, the glans penis should be cleansed thoroughly before specimen collection. The initial few milliliters of urine should be passed into the urinal or toilet bowl to flush out bacteria from the urethra. The midstream portion of the urine is then collected into a sterile container.

B. Catheterized urine samples

The patient should be catheterized by the physician or nursing service using proper aseptic technique. The "free flow" from the mouth of the catheter should be collected into a sterile container. Urine collection by catheterization is seldom indicated, except in those cases where catheterization must be done for diagnostic or therapeutic reasons. There is a small risk of infection after urethral catheterization, which varies with the type of patient catheterized. Because of normal urethral colonization with bacteria, it may be difficult to determine whether organisms isolated from catheterized urine samples are of urinary or urethral origin.

C. Indwelling catheters

In patients with chronic indwelling urethral catheters attached to closed drainage, urine for culture is collected from the wall of the catheter at the junction of the drainage tube. After disinfecting the wall of the catheter with a suitable agent, the wall is punctured with a needle attached to a sterile syringe into which the urine is aspirated. The urine should be transferred to a sterile container for transport to the laboratory. The connection between the catheter and the drainage tube should not be broken for specimen collection, nor should material be taken from the drainage bag.

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D. Cystoscopic urine samples

Cystoscopic urine samples are collected in surgery. They should be transported to the laboratory in sterile containers.

E. Suprapubic Aspiration

Suprapubic aspiration is indicated in patients with clinical evidence of infection but in whom bacterial counts in clean voided specimens are low and therefore indeterminate. This may be seen in neonates, young infants, patients in whom catheterization may be contraindicated and those with suspected anaerobic bacteriuria.

5. **Eye Cultures** (Must be collected by doctors or nurses.)

Specimens should be obtained under aseptic conditions and before antibiotic therapy. Cultures should not be taken within 4 hours after irrigation or installation of disinfectant solutions or ophthalmic medications. Medications may contain antibacterial preservatives. Local anesthetics tend to flush organisms from the conjunctival sac.

6. **Ear Cultures** (May be collected by doctors or nurses.)

A swab is inserted into the external auditory canal where a sampling of pus may be obtained or the inner lining may be swabbed. The swab should be removed without touching the exterior of the ear.

7. **Wound Cultures**

Wound Culture (Deep)


This culture (Wound Culture Deep) is to be used for any surgical specimen (except tissue samples), purulence or necrosis from deep subcutaneous sites and samples collected by needle aspiration. If sample is not collected by needle aspiration and a swab is used, then collect as much specimen as possible and use special care to sample the active site of infection to prevent contamination. Inappropriate specimens would be those contaminated with surface material.

Specimens that are collected should be sent to the Laboratory immediately due to the possible presence of anaerobes.

Wound Culture (Superficial)

The surface of cutaneous wounds or decubitus ulcers frequently are colonized with environmental bacteria and swab samples often do not reflect the true cause of the infectious process. For this reason, the most desirable method of collecting cutaneous specimens is by aspirating lobulated purulent material from the depths of the wound with sterile needle and syringe.

If material cannot be obtained with a needle and syringe, then a swab must be used to collect the specimen. After gently separating the wound margins, extend the tip of the swab deep into the wound, taking care not to touch the adjacent skin margins.

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8. Body Fluid Cultures

Body fluids (ex. synovial, thoracentesis, peritoneal, pleural) are collected by the physician and their collection procedures will not be discussed in this procedure. Submit specimen to the lab in a sterile container such as a sterile urine cup. Body fluids should not be transported to the lab thru the pneumatic tube system.

9. Genital Cultures (including Gonorrhoeae Culture)

Genital cultures will be collected by the physician or nurse.

In the female, the best site to obtain a culture is the cervix. An experienced professional should collect this with care. A sterile swab is inserted into the endocervical canal, moved from side to side, then removed. Vaginal cultures can be obtained by swab at this time also.

In the male, a urethral culture is recommended. This is obtained by inserting a thin swab 2-4 cm into the endourethra, gently rotate it, leave it in place for 1-2 seconds and then withdraw.

Note: Genital cultures collected in the Emergency Room will not be tested for Neisseria gonorrhoeae. ER will collect separate specimens for PCR testing that will be sent to a reference laboratory.

10. Device Cultures

These specimens should be sent to the laboratory in a clean, sterile container.

11. Bronchial Culture

Approximately 15-30 ml of fluid should be collected and transported to the laboratory in a clean, sterile container.

12. Blood Culture

Refer to [Blood Culture Procedure](#) for details or call the laboratory.

For orders of Fungal Cultures on blood send to LabCorp.

- a. Order Fungus Culture # 008482
- b. Draw in 10cc Sodium Heparin Tube (located in Microbiology)
- c. Sample must arrive at LabCorp within 72 hours.


NOTE:

The most common fungemia is caused by *Candida albicans* followed by other *Candida* sp which will be picked up by our BD Bactec FX system. A fungus culture on blood may be ordered when a physician has a patient with immunosuppression that will predispose them to a disseminated fungal infection with organisms like *Histoplasma capsulatum* and *Coccidioides immitis*.

13. Cerebrospinal Fluid Culture

The cerebrospinal fluid is collected by a physician after proper disinfection of the skin.

Fluid is commonly collected into three (3) tubes, the third of which (unless otherwise specified by the physician) is selected for culture. A total of 10 ml is usually collected. This sample should then be transported to the lab STAT. CSF specimen must not be transported to the lab thru the pneumatic tube system.

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14. Strep A Only Culture

Throat culture:

The patient is instructed to breathe deeply and the tongue is gently depressed with a tongue depressor. The swab is then extended between the tonsillar pillars and behind the uvula, taking care not to touch the lateral walls of the buccal cavity. Having the patient phonate an “ah” serves to lift the uvula and aids in reducing the gag reflex. The swab should be swept back and forth across the posterior pharynx to obtain an adequate sample.

15. Strep B Only Culture (to be collected by the physician)

Specimens should be collected from a pregnant woman at 35-37 weeks gestation.

Swab the lower vagina (vaginal introitus), followed by the rectum (i.e., insert swab through the anal sphincter) using the same swab. Cervical cultures are not recommended and a speculum should not be used for culture collection.

Place the swab into a STARSWAB II transport medium.

Transport the clearly identified specimen to the laboratory as soon as possible and request a Strep B Culture. Only Group B Strep will be tested with this order.

**Please note on the order and the swab if the patient is allergic to penicillin.


16. Tissue Culture

Tissue should be collected in a clean, sterile container with a small amount of sterile saline. Samples should be transported to the lab IMMEDIATELY. Anaerobic transport media is available in the Microbiology department for tissue specimens that will have delayed transport.

17. Stool Specimens

Stool Bacterial PCR Panel:

1. Need at least one teaspoon of stool.
2. Stool may be collected in a clean container or may be added to an orange Para-Pak. Place stool in the Para-Pak up to the red fill line. Do not overfill. If collection is from a diapered child it can be facilitated by using saran wrap in the diaper to capture the sample which can then be put into the container provided.
3. Avoid mixing sample with urine, water, soap or toilet paper. Toilet paper may be impregnated with barium salts which are inhibitory to some fecal pathogens.
3. Specimens collected in a clean container or in an orange Para-Pak are stable for **24 hours at room temperature** or for **5 days refrigerated**.
4. The sample container must be labeled with the patient’s name, date of birth, date of collection and time of collection.

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Ordering Notes:

The Stool Bacterial PCR Panel includes testing for Salmonella spp., Campylobacter spp. (C. jejuni and C. coli) Shigella spp / Enteroinvasive E. coli (EIEC), and Shiga toxins 1 & 2.

If Yersinia, Vibrio, Aeromonas or Plesiomonas are ordered, the specimen must be sent to LabCorp for testing. Please refer to the LabCorp directory for specimen requirements.

Diagnostic Testing:

- a. The lab will not accept more than two specimens per patient per day without consultation with the pathology department as there is limited yield provided by additional specimens.
- b. The lab will not accept specimens from inpatients for the Stool Bacterial PCR Panel after the third hospital day without prior consultation. An alternative with those patients over six months of age with clinically significant diarrhea and a history of antibiotic exposure would be to suggest testing for Clostridioides difficile.

Stool Parasite PCR Panel:

1. Stool may be collected in a clean container or may be added to a pink Para-Pak. Place stool in the Para-Pak up to the red fill line. Do not overfill.
2. Avoid mixing sample with urine, water, soap or toilet paper.
3. Specimens collected in a clean container or in an orange Para-Pak are stable for **48 hours at room temperature** or for **5 days refrigerated**.
4. The sample container must be labeled with the patient's name, date of birth, date of collection and time of collection.

Ordering Notes:


The Stool Parasite PCR Panel includes testing for Giardia lamblia, Cryptosporidium (C. hominis and C. parvum only) and Entamoeba histolytica.

Diagnostic Testing:

- a. The lab will not accept more than two specimens per patient per day without consultation with the pathology department as there is limited yield provided by additional specimens.
- b. The lab will not accept specimens from inpatients for ova and parasite testing after the third hospital day without prior consultation.

CDT:

1. Need **at least one teaspoon of stool**.
2. Collect stool sample into a clean, dry container that is free from detergent residue. (if collection is from a diapered child it can be facilitated by using "saran wrap" in the diaper to capture the sample which can then be put into the container provided)
3. Sample must be brought to the lab after collection.
4. Samples in a clean container may be stored at **room temperature for 48 hours** or kept **refrigerated for 5 days**.
5. The sample container must have the patient's name, date of birth, date of collection, and time of collection written on the collection container.

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OCCULT BLOOD (GUAIAAC – HEMOCCULT): [PATIENT INSTRUCTIONS](#) AT END OF THIS PROCEDURE.

Stool submitted for GUAIAAC testing should be submitted in a clean container, maintained at room temperature and placed on Hemoccult cards within 2 hours of collection.

Once specimen is placed on the Hemoccult cards, the cards are stable for 14 days.

OCCULT BLOOD (IMMUNOCHEMICAL – HEMOSURE iFOB):

Specimens submitted for Immunochemical Fecal Occult Blood Testing (iFOB, iFIT) should be submitted in clean container and are stable for 24 hours at room temperature or refrigerated.

There are **no dietary or medication restrictions** associated with the Hemosure iFOB test.

Specimens should not be submitted from patients with menstrual bleeding, bleeding hemorrhoids, constipation bleeding, or urinary bleeding.

WBC, STOOL (Lactoferrin – Qualitative):

1. Need **at least one teaspoon** of stool collected in a clean, dry container. (Specimens collected in preservative are **unacceptable** for testing).
2. Specimens are stable at room temperature or refrigerated for **2 weeks**.
3. Specimens from breast fed infants should not be tested with this methodology.
4. The sample container must have the patient's name, date of birth, date of collection, and time of collection written on the collection container.

WBC,STOOL (Infant SWBC – For Breast fed Infants Only):

1. Need **at least one teaspoon** of stool collected in a clean, dry container. (Specimens collected in preservative are **unacceptable** for testing).
2. Specimens are stable at room temperature for **2 hours**.
3. The sample container must have the patient's name, date of birth, date of collection, and time of collection written on the collection container.

ROTAVIRUS TESTING:

1. Send **at least one teaspoon of stool**.
2. Collect specimen as soon as possible after the onset of disease, preferable 3-5 days after onset. Samples collected after eight days may be less reactive.
3. Collect stool sample into a clean, dry container that is free from detergent residue. (if collection is from a diapered child it can be facilitated by using "saran wrap" in the diaper to capture the sample which can then be put into the container provided)
4. The sample must be brought **to the lab within two hours** of obtaining the sample **or placed in a refrigerator for up to 24 hours** if not able to deliver to the lab within two hours.
5. The sample container must have the patient's name, date of birth, date of collection and time of collection written on the collection container.

18. AFB Cultures and Smears

Use a sterile, leak proof, disposable plastic container to collect a specimen before antimicrobial therapy is begun. Swabs of exudate from skin sources are acceptable, otherwise swab specimens should not be submitted. The sample should be delivered to the laboratory in as short a time as practical to avoid overgrowth by contaminants. If the delivery in transport is delayed by more than one hour the specimen should be refrigerated. A minimum of three early morning sputum specimens collected on three separate days is recommended.

19. RSV Testing

Collect a nasopharyngeal sample using a flocked nasopharyngeal swab.

Place this swab in Viral Transport Media or 1.0 ml Normal Saline.

Break the swab at the break point located on the handle. The swabs, saline tubes and viral transport media are available from Microbiology.

Transport to the lab as rapidly as possible.

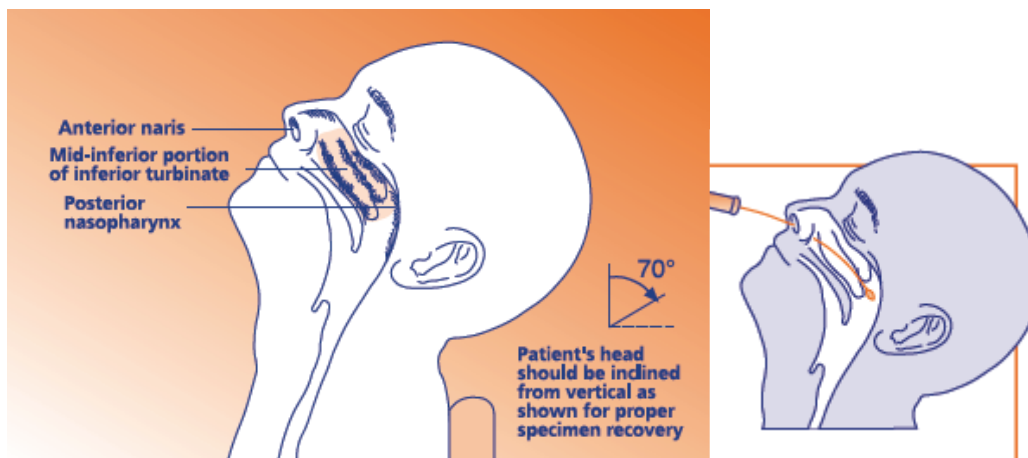
Specimens are stable for 24 hours either at room temperature or refrigerated (2-8°C).


Excessively bloody samples should not be submitted.

20. INFLUENZA A+B Testing

Nasopharyngeal Swab Method

1. Insert flocked nasopharyngeal swab into one nostril. Repeating for the second nostril will deliver optimal combined sample.
2. Rotate swab over surface of posterior nasopharynx.
3. Withdraw swab from collection site and insert into Viral Transport Media or 1 ml. Normal Saline.
4. Transport to the lab as rapidly as possible.
5. Specimens are stable for 4 hours at room temperature and 24 hours refrigerated.



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21. Upper Respiratory PCR Panel (BioFire)

Collect nasopharyngeal specimen using above procedure. Specimen should be collected on a nasopharyngeal swab and placed immediately in viral transport media (M4 or UTM). Swabs and viral transport media can be obtained from the Microbiology laboratory. Specimens are stable at room temperature for 4 hours and refrigerated for 3 days.

22. QUICK STREP (Group A Strep Screen)

Collect specimens with a **Sterile Rayon Swab** (available from Microbiology) from the tonsils and/or the back of the throat. Take care to avoid the teeth, gums, tongue or cheek surfaces.

Place the swab back into the wrapper. Label the wrapper/swab with at least two identifiers (ie. Patient name and date of birth)

Process the swab as soon as possible after collecting the specimen.

23. COVID-19 Testing

PCR Testing

Collect nasopharyngeal specimen using the above procedure. Specimen should be collected on a nasopharyngeal swab and placed immediately in viral transport media (UTM/VTM). Swabs and viral transport media can be obtained from the Microbiology laboratory. Specimens are stable refrigerated for 48 hours.

Antigen Testing


Collect a nasal specimen using a sterile foam tipped applicator. Swabs can be obtained from the Microbiology laboratory. Patient should be instructed to blow their nose prior to collection. Collect the specimen by inserting the swab into the nostril until resistance is met (less than 1 inch into the nostril). Rotate the swab a few times against the nasal wall and remove from nostril. Place swab into original wrapper or container. Do **NOT** place specimen in viral transport media. Testing is performed on the dry swab. Specimens are stable for 48 hours at room temperature or refrigerated.

REFERENCE

Isenberg, Henry D. (2004). Clinical Microbiology Procedures Handbook. (2nd Edition, Vol. 1) Washington DC: ASM Press. Pages 2.0.1 – 2.1.28.

Koneman, Elmer W. (2006). Color Atlas and Textbook of Diagnostic Microbiology. (6th Edition) Baltimore, MD: Lippincott, Williams & Wilkins. Pages 67-110.

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PATIENT INSTRUCTIONS Hemoccult II® BRAND Test Card

CAUTION: For accurate test results, read Patient Instructions completely before collection of sample. Call 419-557-7487 with collection questions.

Drug Guidelines

Please talk to your doctor or pharmacist if you think you have to discontinue a medication for this test.

- For **seven** days before and during the stool collection period **avoid** non-steroidal anti-inflammatory drugs such as ibuprofen, naproxen or aspirin (more than one adult aspirin a day).
- Acetaminophen (Tylenol) can be taken as needed.
- For **three** days before and during the stool collection period **avoid** vitamin C in excess of 250 mg a day from supplements, and citrus fruits and juices.

Diet Guidelines

- For **three** days before and during stool collection period **avoid** red meats (beef, lamb and liver).
- Eat a well balanced diet including fiber such as bran cereals, fruits and vegetables.


Notes:

- Some iron supplements contain vitamin C in excess of 250 mg.
- Do not collect samples if blood is visible in your stool or urine (e.g., menstruation, active hemorrhoids, urinary tract infection). **Contact your doctor.**
- Return completed slide to the laboratory (**do not mail**) no later than 14 days after your first sample collection.
- Protect slide from heat, light, and volatile chemicals (iodine, ammonia, bleach, and household cleaners).

INSTRUCTIONS

1. Using a ball-point pen, write your **name, age, sample collection date, and time** on the front of the card on the lines provided.
2. Use any clean, dry container to collect your sample. Collect sample before it contacts the toilet bowl water. Let stool fall into collection container.
3. **Open front of card** (side where you wrote your name) and use one stick to collect a small sample. **Apply a thin smear** covering Box A.
4. Collect second sample from different part of the same stool with same stick. Apply a **thin smear** covering Box B. Discard stick in a waste container. **DO NOT FLUSH STICK.**
5. Close and secure front flap by inserting it under tab. Store card in any paper envelope.
6. Repeat steps 2-4 for the next two days if there are orders for Occult Blood x3 (**three different days**).

Return to Firelands Regional Medical Center Laboratory

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Obtaining and testing a clean catch (not contaminated) urine specimen is the best way for your doctor to diagnose a urinary tract infection.

INSTRUCTIONS

1. Wash hands with soap and water. Rinse them and dry with paper towel or allow to air dry.
2. Carefully open container. Be sure not to touch the inside of the cap or container.
3. Put the cap on the counter with the inside of the cap face up.
4. Remove the towelettes from the packaging.
5. Cleanse genital area with towelettes as follows:

Females: Using one hand separate folds of urinary opening. With the first towelette, clean one side of the inner fold, using a single *downward* stroke. With the second towelette, clean the opposite side with one *downward* stroke. With the third towelette, clean the center area with one *downward* stroke. Keep folds separated during urination into the container.

Males: Retract the foreskin if present and clean head of penis using one *downward* stroke. Repeat this two more times using a new towelette each time.

6. Begin urination into the toilet. As urination continues, bring container into the “midstream” of the urine to catch the urine in the cup. Fill specimen container only halfway. Finish urinating in the toilet.
7. Without touching the inside of the container or cap, screw the cap on the urine container.
8. The container should be labeled with: your full name, date of birth, today’s date and time of collection. If any of these are missing or incorrect please add or correct as needed.
9. Place the container on the cabinet or shelf.
10. Wash your hands.