

# **BRATTLEBORO MEMORIAL HOSPITAL**

## **2024 LAB GUIDE SPECIMEN COLLECTION**

## SPECIMEN COLLECTION - MISC. NOTES ON SPECIMEN COLLECTION

1. Patient questions concerning the need or significance of tests ordered should be directed to the physician.
2. Steps to take for prevention of hematoma:
  - a) ensure the needle fully penetrates the uppermost wall of the vein.
  - b) remove the tourniquet before removing needle.
  - c) apply slight pressure with gauze when bandaging
3. Steps to take for prevention of hemolysis:
  - a) mix specimens with additives gently but thoroughly
  - b) avoid collecting from a site with a hematoma present
  - c) avoid using a needle that is too small
  - d) make sure the needle is fitted securely to the vacutainer holder
  - e) allow alcohol to dry before venipuncture is performed.
4. Timed specimens must be collected within 30 minutes of scheduled time.
5. Use special, metal free collection tubes for heavy metal tests.
6. Order of draw for multiple specimen collection.
  - 1) First: blood culture tubes
  - 2) Second: non-additive tubes (plain red)
  - 3) Third: coagulation tubes (blue top)
  - 4) Fourth: gel tube (red top with yellow circle)
  - 5) Fifth: additive tubes (green, lavender, gray, other)
7. If drawing with a butterfly for coagulation testing only (PT/PTT/Fibrinogen), draw and discard one 5mL tube first to remove air from the tubing.

**Note:** A blue top tube can be drawn from a central line only if 5 – 10 mL “waste” tube is drawn first to get rid of residual heparin.

### PATIENT CARE: MEDICAL ASSISTANCE

#### **Fainting:**

1. Lower the patient's head or transfer to bed.
2. Loosen tight clothing.
3. Apply cold compresses to forehead and back of neck.
4. Call 111 and ask for a Rapid Response.

#### **Nausea:**

1. Make patient as comfortable as possible.
2. Instruct patient to breathe deeply and slowly.
3. Apply a cold compress to forehead.

#### **Vomiting:**

1. Provide a basin and tissues.
2. Give patient water to rinse out mouth.

#### **Convulsions:**

1. Prevent patient from injuring him/herself.
2. **Do not** restrain patient.
3. Notify the physician for immediate assistance.

**NOTE:** Incident Report must be entered into Incident Reporting system within 24 hours of occurrence.

## SPECIMEN COLLECTION TUBES

Most Laboratory tests are performed on anticoagulated whole blood, plasma, or serum. Specimens should be refrigerated until placed in the courier box for transport to the Laboratory. Please refer to the individual test section for specific requirements. If frozen serum is required, pour off serum into a plastic vial and freeze. Do not freeze collection tubes.

**Special Collection Tubes:** Some tests require specific tubes for proper analysis. Please contact the Laboratory prior to patient draw to obtain the correct tubes for metal analysis or other test as identified in the individual test listings.

**Green Top Tube (Lithium Heparin):** This tube contains Lithium Heparin used for the collection of heparinized plasma or whole blood for special tests. **NOTE: After the tube has been filled, immediately invert the tube several times in order to prevent coagulation.**

**Gray Top Tube (Potassium Oxalate / Sodium Fluoride):** This tube utilizes potassium oxalate as an anticoagulant and sodium fluoride to preserve lactic acid in whole blood and for some special chemistry tests. **NOTE: It is critical that the tube be completely filled. The ratio of blood to anticoagulant is critical for valid test time results. The blood will stop flowing into the tube when adequate volume is obtained. Invert the tube six to ten times immediately after collection to activate the anticoagulant.** Centrifuge and separate within 15 minutes of collection.

**Lavender Top Tube (EDTA):** This tube contains EDTA as an anticoagulant and is used for most hematological procedures and a few special chemistry procedures: **NOTE: After the tube has been filled with blood, immediately invert the tube several times in order to prevent coagulation.**

**Pink Top Tube (EDTA):** This tube contains EDTA as an anticoagulant and is used for most blood bank tests. **NOTE: After the tube has been filled with blood, immediately invert the tube several times in order to prevent coagulation.**

**Light Blue Top Tube (Sodium Citrate):** This tube contains sodium citrate as an anticoagulant used for collection of blood for coagulation studies. **NOTE: It is critical that the tube be completely filled. The ratio of blood to anticoagulant is critical for valid test results. The blood will stop flowing into the tube when adequate volume is obtained.** Invert the tube six to ten times immediately after collection to activate the anticoagulant.

**Red Top Tube:** This tube does not contain any anticoagulant. It is used for collection of serum for selected chemistry and blood bank tests.

**Serum Separator Tube (Red Top with Yellow Circle):** This tube contains a clot activator and serum gel separator used for various serum chemistry tests. **NOTE: Invert the tube to activate the clotting; let stand for 20-30 minutes before centrifuging for 10 minutes.**

**Plasma Separator Tube (Green Top with Yellow Circle):** This tube contains plasma gel separator used for various chemistry tests. **NOTE: Invert the tube about 5 times before centrifuging.**



ACETAMINOPHEN  
 ALBUMIN  
 ALKALINE PHOSPHATASE  
 AMYLASE-  
 BASIC METABOLIC PROFILE  
 CARBAMAZEPINE  
 CARDIAC PROFILE  
 COMPREHENSIVE METABOLIC PROFILE  
 CREATINE KINASE (CK)  
 C-REACTIVE PROTEIN ((CRP)  
 C-REACTIVE PROTIEN, high sensitivity  
 CORTISOL  
 DIGOXIN  
 DIRECT BILIRUBIN (DBIL)  
 ESTRADIOL  
 ETHANOL  
 FERRITIN  
 FREE T3  
 FREE T4  
 FOLLICLE STIMULATING HORMONE (FSH)  
 GAMMA GLUTAMATE TRANSFERRIN (GGT)  
 GENTAMICIN  
 GLUCOSE  
 HORMONE CHORIONIC GONADOTROPIN (HCG)  
 IRON

LACTATE DEHYDROGENASE (LDH)  
 LIPASE  
 LIVER PANEL  
 LUTEINIZING HORMONE (LH)  
 MAGNESIUM  
 PARATHYROID HORMONE (PTH)  
 PHOSPHATE  
 PRO NT BNP  
 SALICYLATE  
 THYROID STIMULATING HORMONE  
 THEOPHYLLINE  
 TROPONIN  
 TOBRAMYCIN  
 TOTAL T3  
 TOTAL T4  
 TOTAL BILIRUBIN  
 URIC ACID  
 PROSTATE SPECIFIC ANTIGEN (PSA)  
 PHENYTOIN (DILATIN)  
 VALPROIC ACID  
 VANCOMYCIN  
 VITAMIN D 25-OH

**NOTE**  
**NO FOLATE, LITHIUM, TIBC OR VIT B12**



### Blood Bank Specimens

(Special labeling sometimes required; please contact lab at 257-8311)

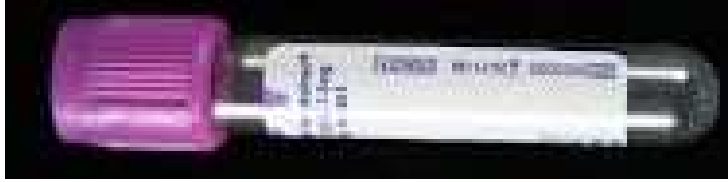



ACETAMINOPHEN  
 ALBUMIN  
 ALKALINE PHOSPHATASE  
 AMYLASE-  
 BASIC METABOLIC PROFILE  
 CARBAMAZEPINE  
 CARDIAC PROFILE  
 COMPREHENSIVE METABOLIC PROFILE  
 CREATINE KINASE (CK)  
 C-REACTIVE PROTEIN ((CRP)  
 C-REACTIVE PROTIEIN, high sensitivity  
 CORTISOL  
 DIGOXIN  
 DIRECT BILIRUBIN (DBIL)  
 ESTRADIOL  
 ETHANOL  
 FERRITIN  
 FREE T3  
 FREE T4  
 FOLATE  
 FOLLICLE STIMULATING HORMONE (FSH)  
 GAMMA GLUTAMATE TRANSFERRIN (GGT)  
 GENTAMICIN  
 GLUCOSE  
 HEPATITIS (A, B, C)  
 HORMONE CHORIONIC GONADOTROPIN (HCG)

IRON  
 LACTATE DEHYDROGENASE (LDH)  
 LIPASE  
 LITHIUM  
 LIVER PANEL  
 LUTEINIZING HORMONE (LH)  
 MAGNESIUM  
 PARATHYROID HORMONE (PTH)  
 PHOSPHATE  
 PRO NT BNP  
 RUBELLA  
 SALICYLATE  
 THYROID STIMULATING HORMONE  
 THEOPHYLLINE  
 TOBRAMYCIN  
 TOTAL T3  
 TOTAL T4  
 TOTAL BILIRUBIN  
 URIC ACID  
 PROSTATE SPECIFIC ANTIGEN (PSA)  
 PHENYTOIN (DILATIN)  
 TOTAL IRON BINDING CAPACITY (TIBC)  
 VALPROIC ACID  
 VANCOMYCIN  
 VITAMIN B12  
 VITAMIN D 25-OH



**Lactic Acid**  
**(Centrifuged and separated within 15 minutes of draw)**



Ammonia on   
Complete Blood Count (CBC)  
Hemoglobin A1c

## AMNIOTIC FLUID

### General Procedure:

- 1) Contact the Laboratory at 802-257-8311.

## **BIOPSIES** **(Tissue other than Bone Marrow)**

### **General Procedure:**

- 1) The specimen(s) submitted for routine light microscopy are to be placed promptly in 10% Formalin which is obtained in small bottles from the Pathology Section of the Laboratory.
- 2) The specimen(s) submitted for additional testing including chromosome analysis or flow cytometry are to be submitted either in sterile saline or dry.
- 3) All specimen(s) requesting "Frozen Section" are to be submitted dry.
- 4) Properly labeled specimen(s) are forwarded to the Laboratory with the appropriate requisition forms.



## BLOOD COLLECTION TUBES (2 Pages)

Stopper Color	Volume Draw	Specimen	Additive / Preservative
Red	8 ml	Serum Clotted Blood	Sterile Interior. No additive. Silicone Coated Interior. Silicone lubricated stopper.
Red	4.0ml	Serum Clotted Blood	Clot activator. Interior coating: Silicone. Silicone lubricated stopper.
Navy / Royal Blue w/ Blue Label	7ml	Plasma Whole Blood	Na <sub>2</sub> EDTA 10.5 Trace Element determinations. No interior coating. Silicone lubricated stopper.
Green	4ml	Plasma Whole Blood	Sterile Interior. Net contents per tube: 45 USP unit of Lithium or Sodium Heparin. No interior coating. Silicone lubricated stopper.
Grey	4ml	Plasma Whole Blood	Sodium Fluoride 10 mg. No interior coating. Potassium Oxalate 8mg. Silicone lubricated stopper. 68 USPS units.
Bactec (Blue)	6-10 ml	Standard 10 Aerobic / F Culture	Dry, Natural Rubber; Processed water, 40 mL; Soybean-Casein Digest Broth 3.0%; Yeast Extract, 0.3%; Animal Tissue Digest, 0.01% ; Sucrose 0.1%; Hemin, 0.0005%; Menadione, 0.00005%; Pyridoxal HCl (Vitamin B6), 0.001% ; Sodium Bicarbonate, 0.04%; Sodium Polyanetholsulfonate, 0.035%
Bactec (Yellow)	6-10 mL	Standard Anaerobic / F Culture	Dry Natural Rubber; Processed water, 40 ml; Soybean-Casein Digest Broth, 3.0%; Yeast Extract, 0.4%; Animal Tissue Digest, 0.01%; Dextrose, 0.25%; Hemin, 0.0005%; Menadione, 0.00005%; Thiols, 0.10%; Sodium Polyanetholesulfonate, 0.025%
Navy/Royal Blue w/red Label	7 ml	Serum	No additive

## BLOOD COLLECTION TUBES (continued)

Stopper Color	Volume Draw	Specimen	Additive / Preservative
Bactec (Pink)	1-5ml	Peds Plus / F Culture	Processed water, 40 ml; Soybean-Casein Digest Broth, 2.75%; Yeast Extract, 0.25%; Animal Tissue Digest, 0.10%; Sodium Pyruvate, 0.10%; Dextrose, 0.06%; Sucrose, 0.08%; Hemin, 0.005%; Menadione, 0.00005%; Sodium Polyanetholesulfonate, 0.020%; Pyridoxal HCl (Vitamin B6), 0.001%; Nonionic Adsorbing Resin, 10.0%; Cationic Exchange Resin, 0.6%.
Lavender	3ml	Plasma Whole Blood	Sterile Interior. Net content per tube: 0.068ml of 7.5% (K <sub>3</sub> ) EDTA solution (5.1mg). Glycerine lubricated stopper. No interior coating.
Red	10ml	Serum Clotted Blood	Contains: Polymer Barrier Material. Clot activator on Interior Walls. Silicone lubricated stopper
Microtainer Red Top	.25 – 0.50	Serum Clotted Blood	Polymer Gel.
Microtainer-Purple Top	Pediatric (0.25 - 0.50)	Plasma Whole Blood	K <sub>2</sub> EDTA
Blue	4ml	Plasma Whole Blood	9 NC Coagulation Sodium Citrate 3.2%
Pink	6.0ml	Plasma	K <sub>2</sub> EDTA 10.8 mg. Silicone lubricated stopper.
Navy/Royal Blue w/ Blue Label	6.0ml	Plasma	Trace Element K <sub>2</sub> EDTA 10.8 mg

## **BLOOD CULTURE COLLECTION (2 pages)**

At least 2 sets of blood cultures are recommended, with a set collected simultaneously from each lumen of an existing central venous catheter (CVC), if present, and from a peripheral vein site; 2 blood culture sets from separate venipunctures should be sent if no central catheter is present.” The Infectious Disease Society of America (2012).

### **DIRECT DRAW OPTIONS**

A. BUTTERFLY: Direct draw into blood culture bottles with vacutainer adapter on bottle. Use marks on bottle to judge blood volume.

B. SYRINGE: Needle with transfer to blood culture bottles.

VOLUME REQUIRED: Anaerobic (yellow cap) 10 mL (Draw this bottle first)  
Aerobic (blue cap) 10 mL

BOTTLE TOP PREP: Wipe rubber top with 70% alcohol pad.

SKIN PREP: Prevantics swabstick Chlorhexidine gluconate (3.15%)- 1-2 minutes skin contact minimum before venipuncture.

### **PEDIATRIC BLOOD CULTURE PROCEDURE**

VOLUME NEEDED: Optimal 1-2 mL  
Minimum 0.5 mL

SAMPLE COLLECTION: Syringe  
Butterfly blood collection set - Direct draw through tubing to Peds Plus culture bottle.  
USE MARKS ON BOTTLE TO JUDGE VOLUME.

SKIN PREP: Providine-Iodine Prep Pad

BOTTLE TOP PREP: Remove flip top cap. Wipe rubber stopper with alcohol. Keep alcohol pad on cap top until ready to inoculate.

AGE: Less than 2 months of age.

## BLOOD CULTURE COLLECTION (continued)

*A volume of 10 mL must be added to each adult blood culture bottle*

*A volume of 3 to 5 mL is recommended for pediatric patients and as little as 0.5 to 1mL for newborns*

Age	Draw Volumes	Instructions
Neonates to 3 years	Draw approximately 1ml blood for each year of life	Transfer entire amount to Pediatric BACTEC bottle
4 years to 10 years	Draw approximately 1ml blood for each year of life	Transfer entire amount to aerobic BACTEC bottle
10 years or over	20 ml	BACTEC aerobic bottle (10 ml) BACTEC anaerobic bottle (10 ml)
Difficult draw 10 years or over	10-20 mL	BACTEC aerobic bottle (10 ml) Remaining blood in BACTEC anaerobic bottle
Difficult draw 10 years or over	Less than 10 mL obtainable	Transfer entire amount to aerobic BACTEC bottle

## BODY FLUIDS COLLECTION (2 Pages)

**AMNIOTIC FETAL LUNG MATURITY** - Uncontaminated amniotic fluid.

**CERVICOVAGINAL SECRETION** - Fetal Fibronectin should only be collected with an Adeza Biomedical Specimen Collection Kit

### PLEURAL / PERICARDIAL

**Container Kits are available from the Lab or else, use as follows:**

- Cell Count - One purple top vacutainer tube.
- Microbiology - 10-50 mL fluid in a sterile red top vacutainer tube, sterile urine cup or a sterile capped syringe (**No needles**).
- Chemistry - One to two red top transfer tubes.
- Cytology - Fresh fluid (>10 mL) submitted in a sterile container.

**Note:** If <10 mL fluid available, call Lab at 257-8311 for instructions.

### PERITONEAL / ASCITES / PARACENTESIS

- Cell Count - One purple top vacutainer tube.
- Microbiology - 10-50 mL fluid in a sterile red top vacutainer tube, or a sterile urine cup.
- Chemistry - One red top and one green top vacutainer tube.
- Cytology - Fresh fluid (10 - 50 mL) submitted in a sterile container.

### SYNOVIAL / JOINT FLUID

- Cell Count and/or Crystals - One purple top vacutainer tube.
- Microbiology - 0.5-10 mL ( $\geq 10$  mL preferred) fluid in a sterile red top vacutainer tube, sterile urine cup or a sterile capped syringe.
- Chemistry - One to two red top transfer tubes for sendout testing.

### VAGINAL FLUID FOR AMNISURE TEST

#### **Patient Preparation in clinician's office:**

NOTE: Until the diagnosis of membrane rupture is excluded, avoid digital cervical examination to prevent infection and shorten the latency period.

- Identify patient according to patient identification policy.
- Position patient flat on back.
- Collect sample of vaginal secretions using sterile vaginal swab provided in kit.
- Remove swab from packaging using care not to touch anything prior to insertion into vagina.
- Collect sample from surface of vagina, holding swab in the middle of the stick while patient is lying flat on back.
- Carefully insert the polyester tip of the swab into the vagina until fingers contact the skin no more than 2-3 inches (5-7 cm) deep.
- Withdraw the swab **after 1 minute**.
- Rinse swab after collection in solvent vial for **1 minute**, and dispose of as indicated in test procedure. Send the sample to the lab as soon as possible after collection within 4 hours.

## CEREBROSPINAL FLUID\*

Tube #	Test	Optimal Volume (ml)
1	Cell Count will be done <u>ONLY</u> if RBC Count in Tube #3 is >30	1.0
2	Culture, Gram Stain, Glucose, Total Protein	2.5
3	Cell Count and Differential	1.5

### Optional CSF Orders

(Additional CSF is Required as Indicated Below)

	CSF
AFB Culture and Smear	1.0 ml
Fungal Culture	1.0 ml
Cryptococcus Antigen	1.0 ml
VDRL	0.5 ml
Cytology	3.0 ml
CEA	1.0 ml
Myelin Basic Protein	2.0 ml
Oligoclonal Bands	4.0ml & serum
Protein Electrophoresis	2.0 ml
LDH	1.0 ml
Glutamine	1.0 ml
Chorionic Gonadotropin	1.0 ml
B-Glucuronic	1.0 ml

### Myelogram CSF Orders

First Tube: Glucose, Total Protein

Last Tube: Cell Count and Differential

\*For specimens with low volume, the Laboratory will contact the ordering department to prioritize testing. If this fails, the laboratory will prioritize testing based upon available information.

## CREATININE CLEARANCE

1. The general procedure for collection of urine is followed.
2. Test orders are placed in HIS for creatinine clearance.

**NOTE:** The patient should be advised to observe all dietary restrictions for the analysis to be performed (alcohol, vitamins, specific food and other medication) for at least 24 hours before starting the collection and during the collection period.

It is not necessary to restrict fluids during these collections. Permit the patient to drink fluids to promote good urinary output.

## **GLUCOSE SCREENING – GESTATIONAL (PREGNANT FEMALES)**

### **PATIENT INSTRUCTIONS:**

1. Your healthcare provider has requested that you have a Gestational Glucose Screening Test. This is a blood test that will give your healthcare provider an idea of how well your body breaks down glucose after drinking a glucose solution.
  2. You will be given 50 gms glucose to drink. Drink the glucose within 5 minutes. The phlebotomist will draw your blood one-hour post dose.
- FASTING IS PREFERRED BUT NOT REQUIRED
  - IF YOU FEEL ILL OR VOMIT DURING THE TEST, PLEASE TELL THE PHLEBOTOMIST.
  - PLEASE DO NOT SMOKE DURING THE TEST.
  - PLEASE REMAIN IN OUR WAITING ROOM AND AT REST UNTIL THE ENTIRE TEST PROCEDURE IS COMPLETE.
  - YOU MAY DRINK WATER, BUT HAVE NO OTHER FOOD OR BEVERAGE DURING THE TEST.



## **GLUCOSE TOLERANCE TEST HOUR- GESTATIONAL (PREGNANT FEMALE)**

### **PATIENT INSTRUCTIONS:**

1. Your healthcare provider has requested that you have a Glucose Tolerance Test done. This is a series of blood tests that will give your healthcare provider an idea of how well your body breaks down glucose after drinking a glucose solution.
  2. Prior to being given glucose to drink, the phlebotomist will draw your blood and send it to the lab to be analyzed.
  3. When the result is called to the phlebotomist, and if the value is acceptable, you will then be given 100 gms of a glucose solution to drink within 5 minutes.
  4. Three more blood glucose levels will be drawn at one hour, two hours and three hours after ingesting the 100 gms oral glucose solution.
- 
- PLEASE BE FASTING FOR AT LEAST 12 HRS. (YOU MAY ONLY DRINK WATER DURING THE FAST)
  - IF YOU FEEL ILL OR VOMIT DURING THE TEST, PLEASE TELL THE PHLEBOTOMIST.
  - PLEASE DO NOT SMOKE DURING THE TEST.
  - PLEASE REMAIN IN OUR WAITING ROOM AND AT REST UNTIL THE ENTIRE TEST PROCEDURE IS COMPLETE.
  - YOU MAY DRINK WATER, BUT HAVE NO OTHER FOOD OR BEVERAGE DURING THE TEST.

## GLUCOSE TOLERANCE TEST - STANDARD ORAL (NON PREGNANT)

### **PATIENT INSTRUCTIONS:**

1. Your healthcare provider has requested that you have a Glucose Tolerance Test done. This is a series of blood tests that will give your healthcare provider an idea of how well your body breaks down glucose after drinking a glucose solution.
  2. Prior to being given 75 gms glucose to drink, the phlebotomist will draw your blood and send it to the lab to be analyzed.
  3. When the result is called to the phlebotomist, and if the value is acceptable, you will then be given 75 gms of a glucose solution to drink within 5 minutes.
  4. One more blood glucose level will be drawn one hour after you drink the solution.
- 
- PLEASE BE FASTING FOR AT LEAST 12 HRS. (YOU MAY ONLY DRINK WATER DURING THE FAST)
  - IF YOU FEEL ILL OR VOMIT DURING THE TEST, PLEASE TELL THE PHLEBOTOMIST.
  - PLEASE DO NOT SMOKE DURING THE TEST.
  - PLEASE REMAIN IN OUR WAITING ROOM AND AT REST UNTIL THE ENTIRE TEST PROCEDURE IS COMPLETE.
  - YOU MAY DRINK WATER, BUT HAVE NO OTHER FOOD OR BEVERAGE DURING THE TEST.

## GLUCOSE (ORAL) & GESTATIONAL TOLERANCE- BMH STAFF INSTRUCTIONS

GESTATIONAL TOLERANCE AND ORAL GLUCOSE TOLERANCE  
(PREGNANT) 100 GMS (NON-PREGNANT) 75  
GMS

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**\* PATIENT MAY NOT LEAVE WAITING ROOM DURING ANY GLUCOSE TOLERANCE OR SCREENING \***

### **A.) GESTATIONAL AND STANDARD ORAL GLUCOSE TOLERANCE**

**BEFORE THE FASTING SPECIMEN IS DRAWN, THE PHLEBOTOMIST SHOULD VERIFY THAT:**

1. THE PATIENT THAT PRESENTS FOR THE FULL TOLERANCE TEST HAS FASTED FOR AT LEAST 12 HOURS BEFORE THE TEST. ONLY WATER CAN BE CONSUMED DURING THE FAST.
2. THE PATIENT UNDERSTANDS HE/SHE SHOULD REMAIN AT REST DURING THE TEST AND REFRAIN FROM SMOKING.
3. THE PATIENT UNDERSTANDS THAT HE/SHE MAY DRINK WATER BUT CAN HAVE NO OTHER FOOD OR BEVERAGES DURING THE TEST.
4. THE PATIENT UNDERSTANDS THAT SUBSEQUENT SPECIMEN COLLECTION WILL BE NECESSARY AND THE PATIENT IS TOLD THE TIME FOR SUBSEQUENT SPECIMEN COLLECTION.

### **B.) GESTATIONAL SCREENING ONLY (PREGNANT)**

- THE PATIENT THAT PRESENTS FOR THE SCREENING TEST WILL BE GIVEN A 50 GRAM GLUCOSE LOAD WITHOUT REGARD TO THE TIME OF THE LAST MEAL OR THE TIME OF DAY. ALTHOUGH FASTING IS PREFERRED.

# MICROBIOLOGY: GENERAL COLLECTION AND HANDLING GUIDE FOR SPECIMENS

Clinical Microbiology attempts to provide pertinent information about microorganisms in a given specimen. Due to the ubiquitous nature of microorganisms, the clinical microbiologist must interpret the recovery of microorganisms in terms of the history and symptomatology of a patient. They must decide which microorganism isolated may be involved in the disease process.

The success of these efforts is dependent on the awareness of the clinician to know:

- what type of specimen is required
- how to obtain that specimen without compromising its microbiological integrity
- the proper delivery of that specimen to the lab

The proper collection of specimens for testing in the Microbiology lab will directly impact on the results obtained. The technique and skills microbiologists have in processing and examining specimens is meaningless if the procurement and transport are not appropriate.

## B. Safety Considerations:

It is extremely important that those who are engaged in the procurement of a specimen be knowledgeable in the proper techniques for collection. This is to ensure that the specimen integrity is maintained and to minimize any risk of exposure to the personnel involved in the collection process. The use of personal protective equipment should be used.

- Gloves should be worn when obtaining or handling specimens.
- Safety glasses should be worn if there is any possibility of aerosols or splashes.
- Protective clothing (gowns, aprons, lab coats) should be worn when doing any procedures that involve obtaining or handling specimens due to the possible exposure.

## C. Labeling and Transport:

Specimens should be placed in the proper transport media, containers, tubes or vials, and sealed to prevent leakage and contamination. Care should be taken to avoid contaminating the outside of the containers.

### **Proper labeling of the specimens is required for processing.**

Inpatients: specimens should be labeled with

Patient's name	Location	Time and date specimen collected
Hospital # or DOB	Specimen source	Doctor's name

All specimens must be placed in a specimen transport bag with the order request form placed in the outside pocket. If the HIS is down a specimen down time form should be used with the with all the information filled in.

Outpatients: specimens should be labeled with

Patient's name	Time and date specimen collected
Specimen source	DOB if possible

All specimens should be placed in a specimen transport bag with the walk-in slip containing the written orders placed in the outside pocket of the plastic bag. Transportation to the lab should be within proper time limits depending on the specimen. This is to avoid the loss of specimen

integrity which can result in the death of potential pathogenic organisms that might be present, or the overgrowth of normal flora which can prevent the isolation of the potential pathogens.

If the specimens must be stored for a period of time before delivery to the lab, proper temperatures should be adhered to. Some specimens should not be refrigerated, and if maintained at room temperature it should not be near any heaters or cold spots. Any refrigerators used for storage should have an internal thermometer to monitor the temperatures (2-8 C range). SPECIMENS SHOULD NOT BE FROZEN.

D. Policy for dealing with unacceptable specimens submitted to the Microbiology department

1. Specimens not labeled with the patient name or any specimen that is mislabeled will be rejected. The nurse or physician will be required to collect another specimen that is labeled correctly.
2. Specimens not collected properly so as to compromise the results will be rejected.
  - Transportation times must be followed to prevent the overgrowth of normal bacterial flora (e.g. sputum, urine, stool). Transport times must be followed in certain specimens to prevent the loss of fastidious, fragile organisms that may not be recovered on culture if delays occur (e.g. CSF, body fluids, tissue).
  - The proper containers must be used to collect and transport the specimen so organisms are not exposed to conditions that may prevent recovery on culture and to avoid contamination.
  - The specimens should be received in the lab in a condition that will not compromise its integrity and will not expose the lab to unnecessary hazards.
  - There must be adequate volume for certain testing to be performed. If the volume is such that the chance of recovering potential pathogens has been compromised the specimen will be rejected.

If specimens have been handled in a manner unacceptable to obtaining valid results, the nurse or physician will be required to collect another specimen. If this is not possible the nurse must notify the doctor. If the doctor still requests the specimen be processed after being informed of the problem, the Laboratory Director or Pathologist must be notified. The lab will include in the report the results may be compromised due to the improper collection of the specimen.

References:

1. Manual of Clinical Microbiology, 11<sup>th</sup> edition, 2015.
2. Laboratory Diagnosis of Infectious Diseases Principles and Practice, 1st edition, 1988
3. ASM Clinical Microbiology Procedures Handbook. 3<sup>rd</sup> ed. 2010. Section 2.

## **SPECIMENS TO BE CULTURED ANAEROBICALLY**

As a general rule, anaerobic cultures should be obtained from areas of the body not likely to be contaminated with normal flora.

1. Blood
2. Deep wounds, abscesses, aspirated pus
3. Body fluids (pleural, paracentesis, pericardial, synovial, peritoneal, bile, etc)
4. Bone marrow
5. Tissue (biopsy, surgical, autopsy)
6. Transtracheal aspirates, direct lung aspirates

## **SPECIMENS NOT TO BE CULTURED ANAEROBICALLY**

In general specimens from areas with anaerobes as part of the normal flora, or specimens that cannot be obtained without contamination by the normal anaerobic flora should not be cultured anaerobically.

1. Throat, gingival, nasopharyngeal
2. Expecterated sputum
3. Bronchoscopic specimens not collected by protective double lumen catheter
4. Gastric contents, small bowel contents, feces, rectal swabs, colocutaneous fistulae and colostomy stomata
5. Surface material from decubitus ulcers, swab samples of other superficial areas sinus tracts, exchars
6. Material adjacent to skin or mucous membranes which have not been properly decontaminated.
7. Voided urine
8. Vaginal or cervical swabs

## **COLLECTION OF SPECIMENS FOR ANAEROBIC CULTURE**

**Tissue or fluid obtained from a site presumed to be involved in the disease process is always superior to swab specimens. Swab specimens should not be collected when a more representative specimen such as tissue or fluid can be obtained.**

**Tissue:** place a representative portion of tissue (in general the more specimen the better) in a sterile cup. Transport ASAP to the Microbiology department for immediate processing.

**Aspirated fluid:** When aspirating fluid (pus) be careful to minimize air entering the syringe, and expel any excess air before recapping (always using the one handed technique to avoid needle sticks). Place in a plastic bag and transport immediately. The lab can perform (depending on the orders) aerobic, anaerobic, TB and fungal cultures including smears on fluids (if enough volume is received).

**Swab specimen:** collect a swab specimen only when tissue or fluid is unavailable. Swabs should be used to extensively sample a representative portion.

- a. For aerobic culture: the routine aerobic swab should be used
- b. For anaerobic culture: an anaerobic transport swab must be used. It comes in a sterile pack

- Peel package apart, remove swab envelope and peel apart to get at the sterile swabs
- Obtain specimen using the sterile swabs (sample extensively)
- Using sterile technique, remove the tube cap and immediately insert swab down into the medium to within approximately 5mm from the bottom of the tube. Break off the wooden shaft from the swabs that remain outside the tube so the cap can be screwed back on tightly.
- transport to lab within 8-24 hours.

### **Miscellaneous Arthropod ID**

Organisms to be submitted for identification should be placed in a clean cup with a little saline added so they do not dry out (bed bugs, ticks, fleas, lice).

Lice found on hair can be submitted by cutting a piece of the hair which has the adults, nymphs and nits and placing in a container (no additives required).

### **Mites (Sarcoptes scabiei)**

Mites that cause scabies are usually located in a papule or vesicle in the epidermis or tiny burrow in the superficial skin. To obtain specimen for identification:

- tease the mite from its burrow or papule with a needle; or cut a tiny bit of skin with a razor blade and mount on a slide with a cover slip

scrape the skin with a scalpel (can use a drop of mineral oil). It may require searching along the area of inflammation and actually scraping down to cause bleeding to find the mite, usually a female with an egg.

## **HAIR, SKIN, AND NAILS FOR FUNGAL CULTURE - COLLECTION INSTRUCTIONS**

**The diagnosis of fungal infections is dependent upon the proper collection and transport of specimens to the clinical laboratory. The following instructions are for the collection of hair, skin and nails for culture.**

### **MATERIALS NEEDED:**

Gloves	70% alcohol
Scalpel blade or microscope slide	Sterile Container
Forceps	Slide Holder

### **COLLECTION OF HAIR FOR CULTURE:**

- Put on gloves.
- With forceps, collect at least 10 to 12 affected hairs with base shaft intact.
- Place hairs in a sterile container.
- Transport to lab at room temperature within 24 hours of collection.

### **COLLECTION OF NAILS FOR CULTURE:**

- Put on gloves.
- Wipe the nail off with 70% alcohol using gauze (not cotton balls).
- Clip away a generous portion of the affected nail and collect material or debris from under the nail. (Minimum volume: enough material to cover the head of a thumbtack)
- Place material in a sterile container.
- Transport to lab at room temperature within 24 hours of collection.

**COLLECTION OF SKIN FOR CULTURE:**

1. Put on gloves.
2. Clean the affected area with 70% alcohol using gauze (not cotton balls).
3. Gently scrape the surface of the skin at the active margin of the lesion using a scalpel blade or microscope slide. **Do not draw blood!** (Minimum volume: enough material to cover the head of a thumbtack)
4. Collect the scrapings in a sterile container. If specimen is to be submitted between glass slides, tape the slides together and place in a slide holder.
5. Transport to lab at room temperature within 24 hours of collection.

**SAFETY NOTE:** Scalpel blade should be properly discarded in a “sharps” container. **Do not** send blade to the lab.



**COLLECTION AND HANDLING PROCEDURES FOR MICROBIOLOGY SPECIMENS**

<b>SPECIMEN / TEST</b>	<b>TRANSPORT / TIME</b>	<b>CONTAINER / REQUIREMENTS</b>
<b>ANAEROBIC CULTURES</b>	see page <b>XXX</b> for details and specific sites to culture anaerobically	
<b>BLOOD CULTURES</b>	blood is drawn directly into 2 bottles containing 100 ml or 50 ml or media	adult: volume = 10ml per bottle, with 2 bottles drawn per order (set), 100 ml bottles are used for adults. Infants (newborns): 1-5 ml drawn into 50ml bottles
Venipuncture site must be cleansed with 95% alcohol followed by cleansing with 2% iodine. Stoppers of the blood culture bottles should be cleaned with alcohol.		
a. Two sets are drawn automatically on all adult blood culture orders b. If more cultures are required or different timing intervals – the physician must specify.		
<b>CATHETER CULTURE</b>	within 15 minutes	a 2 inch distal segment of catheter should be obtained by aseptically clipping off end directly into sterile container.
<b>URINARY CATHETERS ARE UNACCEPTABLE</b>		
<b>CHLAMYDIA CULTURE</b>	Requires direct inoculation into transport vial - MUST CALL MICRO LAB FOR MEDIA.	
<b>CHLAMYDIA DNA PROBE</b>	within 5 days at room temp.	MUST USE SWAB IN KIT. See Section on N. GONORRHOEAE AND/OR C. TRACHOMATIS TESTING for details.
<b>CLOSTRIDIODES DIFFICILE</b>	within 1-2 hrs if at room temp. within 8-24 hrs if refrigerated	Requires fresh stool (about 1/4 cup). Stool in transport media is unacceptable
A liquid specimen should be submitted. Patient should be producing multiple liquid specimens per day before testing for toxin is done. An antigen screen and toxin test is performed. Both Results must be correlated with patient symptoms.		
<b>COVID-19 (SARS-COV-2)</b>	Within 24 hours refrigerated	Must use swab/media provided by the lab.
<b>CSF CULTURE/GRAM STAIN</b>	immediate	1-5ml fluid sent in a sterile capped tube
<b>TB CULTURE</b>	immediate	requires 5ml of CSF for AFB culture and smear unacceptable if inadequate volume
<b>FUNGAL</b>	immediate	1-5ml fluid sent in a sterile capped tube
Lumbar puncture must be performed using strict asepsis, since contamination of the specimen can occur readily and confuse the identification of the etiologic agent. Skin should be disinfected with povidone-iodine. Sterile screw capped tubes must be sealed well to avoid leakage and contamination.		
Viral meningoencephalitis is frequently established by exclusion and by serology. Some viruses may be isolated from CSF, however viral isolates are more likely to be obtained from other sources.		

SPECIMEN / TEST	TRANSPORT / TIME	CONTAINER / REQUIREMENTS
<b>EAR CULTURE</b>	within 8 hours within 1-2 hours ASAP	routine aerobic culture swab mini-tip aerobic culture swab tympanocentesis, send fluid in syringe directly to lab
Acute otitis media in children often does not require culture because the bacteriological findings have been sufficiently consistent in reported studies. Tympanocentesis may be considered in neonates and elderly, since etiology of otitis media in these populations is unpredictable. Cultures of the nasopharynx or pharynx are not useful for establishing the cause of middle ear infections and should not be done for this purpose.		
<b>EYE CULTURE</b>	within 30 minutes	Aerobic culture swab
<b>SCRAPINGS</b>	<b>STAT</b>	Requires STAT delivery, or inoculation directly on media. Call the Micro lab
<b>GRAM STAIN</b>	no time limit	apply material directly to slide, air dry
It is recommended that swabs for culture be taken before topical anesthetic		
<b>EXUDATES/ASPIRATES</b>	see " wound cultures"	
<b>FECES CULTURE</b>	within 3 days	Stool in Cary Blair transport media. Multiple specs (2-3) should be spaced out (at least a day apart)
<b>RECTAL SWAB</b>	within 1-2 hours	May be used only when passed feces cannot be obtained. The aerobic culture swab should be passed beyond the anal sphincter, carefully rotate, and withdraw
<b>FECES IS UNACCEPTABLE IF CONTAMINATED WITH URINE OR IN A DIAPER</b>		
<b>FUNGAL CULTURE</b>	same requirements as for the routine culture in Cary Blair transport media)	
<b>N.GONORRHOEAE CULTURE</b>	ASAP	Rectal swab is required that must be plated directly on a Jembec plate, Call Micro lab for plates. The swab should be passed beyond the anal sphincter, carefully rotate and withdraw. If contaminated with feces collect another specimen not contaminated with feces.
<b>OVA AND PARASITE</b>	no time limit	feces collected in Total Fix transport vial
<b>RECOMMEND 2 TO 3 SPECIMENS AT LEAST A DAY APART (PREFER SPACED OUT OVER 1 WEEK) SWABS ARE UNACCEPTABLE. SPECIMEN UNACCEPTABLE IF CONTAMINATED WITH URINE, BARIUM,DYES, IRON, BISMUTH, MINERAL OR CASTOR OILS, OR ANTIMICROBICS. MUST ALLOW 5-10 DAYSTO CLEAR IF ABOVE TREATMENTS ARE USED BEFORE COLLECTING SPECIMENS FOR TESTING.</b>		
<b>ROTAVIRUS</b>	within 1 hour	fresh feces in a clean plastic cup unrefrigerated
	8 to 24 hours	fresh feces in a clean plastic cup refrigerated
<b>GIARDIA ANTIGEN</b>	within 48 hours	feces in Cary Blair transport media
	within 7 days	feces in SAF * this is the best collection method.



SPECIMEN / TEST	TRANSPORT / TIME	CONTAINER / REQUIREMENTS
<b>RESPIRATORY TRACT CULTURES</b>		
<b>NASOPHARYNX CULTURE</b>	within 1 hour	minitip N/P culture swab
If culture for <i>C. diphtheriae</i> or Bordetella pertussis - Must call Micro lab. Test is sent to reference laboratory. Offices can order pertussis kits directly from the State lab, and mail in when used.		
<b>NASOPHARYNGEAL WASH</b>	within 8 hours	for Respiratory Syncytial Virus (RSV) only Requires nasal washing that is aspirated up into a syringe.
<b>THROAT</b>	Within 48 hours	routine aerobic culture swab
Beta strep screen: Specimen is cultured for all groups of beta hemolytic Strep (A, B, C, G, F) Routine throat culture: Culture includes screening for all groups of beta hemolytic Strep in addition to screening for a predominance of Haemophilus, S.pneumoniae, or Yeast. For all throat cultures the tongue, gum, cheek or teeth areas should be avoided. Sample the back area of the throat and the tonsillar region.		
<b>SPUTUM CULTURE</b>	within 30 minutes	a fresh, deep cough in a sterile container, automatically includes a gram stain smear
Expectorated sputum is frequently contaminated with oropharyngeal flora, which makes it difficult to determine which of the organisms isolated is responsible for pulmonary infection. All sputa are screened prior to culture. If greater than 25 squamous epithelial cells per low power microscopically are observed then the specimen is rejected and a new one is requested.		
<b>SPUTUM TB CULTURE</b>	within 1 hour	a fresh, deep cough in a sterile container
<b>TB STAIN (AFS)</b>	within 1 hour	a fresh, deep cough in a sterile container
MULTIPLE SPECIMENS SHOULD BE OBTAINED AT DAILY INTERVALS UNLESS OTHERWISE ORDERED BY PHYSICIAN ( CALL THE MICRO LAB )		
<b>BRONCH WASHES/BAL</b>		
<b>ROUTINE CULTURE</b>	immediate	fluid in a sterile container
Gram stain smears are automatically performed		unacceptable for anaerobic culture unless a double lumen catheter is used to eliminate oropharyngeal contamination.
<b>FUNGAL</b>	immediate	fluid in a sterile container
<b>TB CULTURE</b>	immediate	fluid in sterile container
AFB smears are automatically performed		
<b>TRANSTRACHEAL ASPIRATES</b>		
<b>CULTURE/SMEAR</b>	immediate	sterile syringe/trap/or container
<b>ANAEROBIC</b>	immediate	sterile syringe/trap/or container
<b>FUNGAL</b>	immediate	sterile syringe/trap or container
<b>TB CULTURE/SMEAR</b>	immediate	sterile syringe/trap or container

<b>SPECIMEN / TEST</b>	<b>TRANSPORT / TIME</b>	<b>CONTAINER / REQUIREMENTS</b>
<b>PERTUSSIS CULTURES/PCR</b>	ASAP	Pertussis kits, Call Micro lab for Kit or Kits can be ordered directly from the State Laboratory
NP cultures are required and immediate inoculation of media in the kits is necessary. NP cultures not received in the transport media are unacceptable. NP cultures are sent to the State lab.		
<b>SCHLICHTER TEST</b>	MUST CALL BACTI TO ORDER. MICRO LAB MUST HAVE A BACTERIAL ISOLATE FROM THE PATIENT, AND PATIENT MUST BE DRAWN FOR ANTIBIOTIC LEVELS.	
<b>SURGICAL / TISSUE</b>		
<b>CULTURE /SMEARS</b>	immediate	sterile container, Unacceptable if in formalin
<b>FUNGAL</b>	immediate	sterile container, Unacceptable if in formalin
<b>TB CULTURE/SMEAR</b>	immediate	sterile container, Unacceptable if in formalin
<b>TB CULTURES</b>		
<b>RESPIRATORY SPECIMENS - see respiratory tract under specimens</b>		
<b>FLUIDS</b>	immediate	5-10 ml minimum required. More volume increases the chance of recovery
<b>GASTRIC</b>	immediate	Sterile container, requires neutralization, call Micro lab prior to collection. Must be done prior to feeding in the AM.
<b>TISSUE</b>	immediate	sterile container
<b>URINE</b>	within 1 hour	must be clean catch first am specimen in sterile cup or catheter specimen, BD vacutainer tubes are unacceptable
<b>URINE</b>		
<b>CULTURE IN A BD TUBE</b>	within 48 hours	see package for instructions, recommend First AM midstream specimen
<b>CLEAN CATCH (CC)</b>	within 1 hour refrigerate within 2-8 hrs	requires cleaning patient, and obtaining a Midstream specimen in a sterile cup
<b>CATHETER SPECIMEN</b>	within 1 hour if in BD tube within 8 hrs	in syringe or sterile cup, recommend collecting cath urine in BD tube for transport
<b>SUPRAPUBIC ASPIRATE</b>	immediate	specimen is acceptable for routine bacterial culture, TB, fungal and anaerobic culture
<b>GRAM STAIN</b>	within 1 hour	must be a clean catch or cath. BD tube is unacceptable
<b>TB CULTURE</b>	within 1-2 hours	must be first AM clean catch specimen in sterile cup or a catheter specimen

SPECIMEN / TEST	TRANSPORT / TIME	CONTAINER / REQUIREMENTS
<b>WOUNDS/EXUDATES/ASPIRATES/FLUIDS - FOR BODY FLUIDS (PLEURAL, JOINT ETC) SEE BODY FLUIDS</b>		
<b>FLUID,EXUDATE,ASPIRATION</b>		
<b>CULTURE/SMEAR</b>	immediate	in a sterile syringe or tube
Actual fluid/pus is the best specimen to collect for optimum recovery. Aerobic, anaerobic and gram stain smears are automatically performed using special concentration procedures.		
<b>FUNGAL CULTURE</b>	immediate	in a sterile syringe or tube
<b>TB CULTURE</b>	immediate	in a sterile syringe or tube , adequate volume is essential in order to detect small numbers
<b>WOUNDS, ABSCESSSES</b>		
<b>AEROBIC CULTURE</b>	within 8 hours	aerobic culture swab or eswab
<b>GRAM STAIN</b>	within 8 hours	requires a second Aerobic culture swab
<b>ANAEROBIC CULT</b>	within 8 hours	Eswab transport swabs, see package for instructions
<b>FUNGAL CULTURE</b>	within 8 hours	aerobic culture swab - actual fluid or tissue is needed for optimum recovery of fungal pathogens
<b>TB CULTURE</b>	MUST HAVE ACTUAL FLUID OR TISSUE, SWABS UNACCEPTABLE	
<b>MYCOLOGY / FUNGAL CULTURES</b>		
<b>SKIN SCRAPINGS</b>	within 8 hours	sterile plastic container
Area should be cleansed with 70% alcohol swabs. Then scrap the active outer edge of the lesion or area. If topical treatment has occurred, wait at least one week before obtaining specimen.		
<b>HAIR</b>	within 8 hours	sterile container
Remove fluorescing hairs and scrap scales around the follicle. Sample infected hairs broken at hairline by scraping or dig out with dull blade. Send scrapings and infected hair samples.		
<b>NAIL CLIPPINGS</b>	within 8 hours	sterile container
Remove and discard the outer debris, which may contain saprophytes and bacteria. Scrape under nail's dorsal surface until affected area is reached. Submit in container or glass tube. Do not send large pieces of nail if at all possible. For other specimens see individual site procedures		
<b>VIRUS CULTURES</b>		
<b>BODY FLUIDS, CSF, FECES, SPUTUM</b>	immediate	sterile container
<b>THROAT, ULCERS VESICLES</b>	within 1 hour	Viral transport media required (VTM), Call lab for kits see instructions on package for specimen collection
Viral cultures are sent to a reference lab. They can take up to 1 month before completing. If a specific viral agent is suspected, it is important that the requisition state the virus. Methods for detection vary and more rapid results may be possible if specifics are known. Herpes simplex virus should be requested specifically, ordering a Herpes virus culture. This takes up to a week for recovery, positive results are very often called within 48 hours of receiving the specimen. Certain viruses (CMV) require same day shipping to reference laboratory, which requires that the specimen be collected in the morning. It then must be delivered to the laboratory before noon so that it can be shipped that same day. For all viruses it is recommended that the specimens be shipped within 24 hours for greater chance of recovery.		

## N. GONORRHOEAE /C. TRACHOMATIS TESTING/ T. VAGINALIS TESTING

### 1. Endocervical Specimens:

- A. Do not collect specimen at the posterior fornix.
- B. Lukewarm water may be used to warm and lubricate the speculum.
- C. Holding the swab by the white cap, insert the swab into the cervical canal and rotate for 15 to 30 seconds.
- D. Withdraw swab carefully avoiding contact with vaginal surfaces.
- E. Fully insert the swab into the tube so that the tip is at the bottom
- F. Carefully break the shaft at the score mark. Be careful to avoid splashing.
- G. Tightly recap the tube.
- H. Label tube with the patient information, date and time collected. Be careful not to obscure the all the barcodes on the Sample Buffer tube.
- I. Clinician-collected endocervical swab or patient-collected vaginal swab specimens' **must be transferred immediately** (preferred) or within two hours after collection into the BD MAX UVE Sample Buffer Tube when kept at 2 – 30 °C. Protect again exposure to excessive heat.

### 2. Urine Specimens

- A. First void urine specimens **must be transferred** from the collection cup to the BD MAX UVE Sample Buffer Tube within 4 hours of collection when kept at 2 – 30 ° C or within 24 hours of collection when stored at 2 – 8 °C.
- B. Transfer 1 ml of urine into the specimen tube using the provided pipette and invert 3-4 times.
- C. Patient should not have urinated for at least 1 hour prior to specimen collection.
- D. Direct patient to provide first-catch urine (approximately 20-30 ml of initial urine stream) into urine collection cup free of any preservatives. Collection of larger volumes of urine may result in specimen dilution that may reduce test sensitivity.
- E. Female patients should not cleanse labial area prior to providing specimen.
- F. Make sure collection cup cap is closed tightly and labeled with patient name.
- G. Urine specimen is stable for 5 days at room temperature or refrigerated when stored in the UVE Sample Buffer Tube Urine specimen in a sterile container is only stable for 24 hours after collection.

### 3. Preparation for Transport:

- A. Fully insert one BD MAX UVE swab into the BD MAX UVE Sample Buffer Tube.
- B. Snap off shaft at score line or cut shaft to fit tube. Use care to avoid splashing contents.
- C. Cap tube tightly.
- D. For urine, transfer 1 ml of urine into the BD MAX UVE Sample Buffer Tube.
- E. Cap tube tightly. Transport swab tube to the laboratory at 2° to 25°C. Store at 2° to 25°C and test within 5 days. If not tested within 5 days, freeze and store at -20°C for up to 30 days after collection.
- F. Urine sent in a collection cup must be transported to the laboratory at 2° to 30°C, and transferred within 24 hours of collection into the BD MAX UVE Sample Buffer Tube. Store at 2° to 25°C and test within 5 days. If not tested within 5 days, freeze and store at -20°C for up to 30 days after collection.

**NOTE:** Specimens collected with this system cannot be used for culture. Only the approved BD MAX UVE swabs should be used for specimen collection.

Table: Specimen Stability Prior to Transfer into the BD MAX UVE Sample Buffer Tube

Specimen Stability	Specimen Type	Transport and/or Storage Temperature	
		22–30 °C	2–8 °C
Prior to Transfer into BD MAX UVE Sample Buffer Tube	Urine	4 hours	24 hours
	Vaginal/Endocervical Swab	Transfer immediately (preferred) or within 2 hours	
Specimen Stability	Specimen Type	Transport and/or Storage Temperature	
		2–30 °C	-20 °C
In BD MAX UVE Sample Buffer Tube (prior to pre-warm)	Urine and Vaginal/Endocervical Swab	5 days	30 days

4. **For Miscellaneous sites**

- A. Sites: Ocular, Urethral, Oral, or Anal:  
Use Aptima Swab and order Chlam/GC, Misc.
- B. Peritoneal fluid (pelvic wash, cul-de-sac fluid):  
Transfer 1 ml fluid into **the Aptima container** within 24 hours of collection **and order Chlam/GC, Misc**



**GENERAL GUIDE FOR OBTAINING SPECIMENS FOR VIRAL CULTURE**  
**(FOR MORE SPECIFIC INFORMATION, SEE REFERENCE SPECIMEN COLLECTION GUIDE)**

<b>DISEASE CATEGORIES</b>	<b>VIRUSES</b>	<b>SPECIMEN</b>
CENTRAL NERVOUS SYSTEM (aseptic meningitis, encephalitis)	Enterovirus	CSF, feces or rectal swab
	Herpes simplex	Throat, CSF, vesicle fluid, urine
	Arena/Arbo virus (lymphocytic choriomeningitis and equine encephalitis) (Calif, St.Louis, Jap. B encephalitis arboviruses cannot be cultured)	blood, CSF
	Rabies	saliva
CONGENITAL ANOMALIES	Cytomegalovirus	urine, throat
	Rubella virus	CSF, urine
	Herpes simplex	CSF, throat, vesicle aspirate
ENTERITIS stool rotavirus	Rotavirus	feces- Antigen testing performed- see isolation / testing not done
	Norwalk	isolation / testing not done
EXANTHEMATOUS	Measles	throat
	Rubella	throat
	Enterovirus	feces, throat
<b>DISEASE CATEGORIES</b>	<b>VIRUSES</b>	<b>SPECIMEN</b>
CUTANEOUS MUCUS MEMBRANE VESICULAR	Vaccinia, Herpes simplex Varicella zoster	vesicle fluid
	Enterovirus	feces, vesicle fluid, throat
INFECTIOUS MONONUCLEOSIS	Epstein Barr	isolation is not performed
	Cytomegalovirus	urine, throat
RESPIRATORY INFECTIONS (pharyngitis, croup bronchitis, pneumonia)	Adenovirus, CMV, HSV Enterovirus, Rhinovirus Reovirus, Mumps Influenzae, Parainfluenzae Resp.Syncytial virus (RSV) SARS-COV-2 (Covid-19)	throat, sputum, nasal secretions  Antigen test performed, See Nasopharyngeal secretion
Respiratory cultures		
Pleurodynia	Coxsackie virus	throat, feces

## **REFERENCE LABORATORIES (Send-Outs)**

Certain tests are not performed in the BMH Laboratory. They are forwarded to a reference laboratory. These analyses must be requested through the Clinical Laboratory. "Send-out" analysis is routinely sent to pre-selected and approved reference laboratories.

These laboratories are fully accredited by the College of American Pathologists. They are also approved to provide laboratory services under the Medicare Programs and are licensed to operate in interstate commerce by the Center for Disease Control. Consult the Laboratory at (802) 257-8311 for further information.

For specimens sent to reference labs, the Laboratory follows all requisition, collection and handling specifications of the reference laboratory.

## RSV & INFLUENZA A & B ANTIGEN TESTING

1. **Availability:**
  - A. Only order this test on children 5 years and under.
  - B. Outpatients are collected in the physician's office.
  - C. Emergency Department patients are collected by Physician or ED staff.
  - D. Inpatients are collected by Respiratory Therapy staff.
  
2. **Planning:**

Have on hand the following materials:

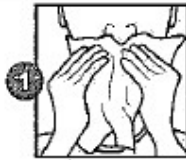
  - A. 3cc syringe.
  - B. 12" butterfly IV tubing.
  - C. Viral transport media (obtained from Microbiology).
  - D. Powder free gloves.
  - E. Laboratory requisition
  - F. Plastic bag with ice in it if delivery to lab is >1 hour.
  - G. Scissors
  
3. **Implementation:**
  - A. Explain procedure to patient.
  - B. Check child's I.D. bracelet.
  - C. Have help on hand if it is necessary to restrain child.
  - D. Take butterfly tubing and cut tubing where it meets the hub of the butterfly.
  - E. Draw up 1cc of N/S into syringe and remove needle.
  - F. Attach the butterfly tubing to syringe.
  - G. Have someone tilt child's head slightly backward.
  - H. Gently insert butterfly tubing in the nostril to nasal pharyngeal area and flush the area with the N/S in the syringe.
  - I. Immediately but gently pull back on the syringe plunger to aspirate the fluid back into the syringe.
  - J. Remove the tubing from the nostril.
  - K. Aspirate transport media into syringe to mix with N/S washing and return all fluid to the transport media container. Make sure cover is secure when completed.
  - L. Label the media vial with patient's name, room number, physician and medical record number. (May use stamper plate).
  - M. Place transport media vial in the plastic bag containing ice and bring it immediately along with the requisition to the Central Processing Area in the laboratory. Delivery to the lab within 1 hour does not require transport on ice.

**NOTE: If Flu is ordered without RSV, nasal or throat collection using a flocced swab (E-swab) will be accepted in lieu of the naso-pharyngeal washing sample.**

## SARS COV-2 (Covid-19) NASAL SPECIMEN COLLECTION

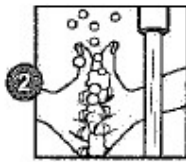
### How to collect an observed nasal swab sample

Read instructions entirely. Failure to follow the instructions entirely may lead to false results. Please only collect the sample in the presence of a staff member in the drop-off station.



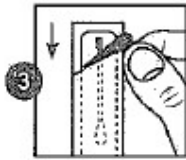
#### 1 Blow your nose.

Make sure it is clear of particulate matter.



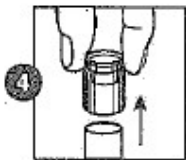
#### 2 Wash your hands.

Wash with soap and water for at least 20 seconds or use hand sanitizer and dry completely.



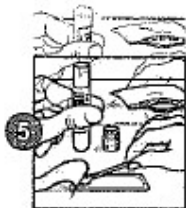
#### 3 Open the package with the swab.

Careful: Don't touch the soft tip with your hands. Peel open where indicated. Leave swab in the package for now.



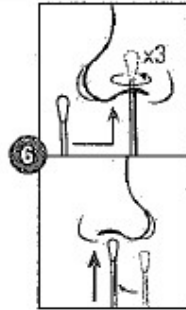
#### 4 Remove the cap of the collection tube.

Place it right side up on a clean surface where you can easily find it.



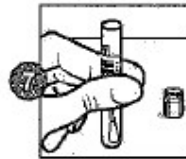
#### 5 Pick up the swab without touching the soft tip.

Have the tube ready to put the swab in after collecting the sample.



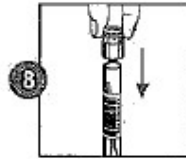
#### 6 Collect sample from both nostrils.

Pull swab out of its packaging, being careful not to touch the soft tip with your hands, and insert it into one nostril just until the soft tip is no longer visible. Rotate it in a circle around the inside edge of your nostril at least 3 times. Use the same soft tip to repeat the previous step in the second nostril 3 times.



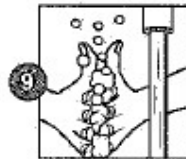
#### 7 Put the swab in the collection tube.

The soft tip of the swab that went into your nose should go into the tube first.



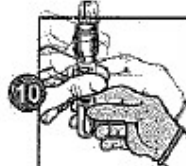
#### 8 Replace the cap.

You're almost done! Make sure the cap is on tight.



#### 9 Wash your hands.

Wash with soap and water for at least 20 seconds or use hand sanitizer and dry completely.



#### 10 Hand tube to staff member.

You are all set!

Questions? Ask a staff member for assistance.

## SEMINAL FLUID ANALYSIS (2Pages)

### SCHEDULING INSTRUCTIONS

1. Obtain a labeled collection container from your physician or the Laboratory.
2. **Call the Laboratory at 257-8311 to make an appointment** for the test. Specimens are accepted for this test **Monday through Friday, 7:00 AM to 12:00 Noon. No weekends or Holidays.**
3. Call 257-8311 to pre-register (this is not required but strongly recommended to save you time).
4. Bring specimen (see collection instructions) directly to the Laboratory in the basement of the hospital.
5. After specimen has been given to the Lab, proceed to Patient Registration. (Even if you have pre-registered, a short visit to this department is necessary to complete the registration process.)

### COLLECTION INSTRUCTIONS

1. Abstain from sexual activity (including masturbation) for 2-3 days (48-72 hours) and no more than 5 days prior to specimen collection.
2. Obtain the semen sample by masturbating directly into a sterile container, provided either by your doctor or the Laboratory. Be sure and collect the **entire** sample. Avoid the use of lubricants to prevent specimen contamination.
3. Put your full name, date of birth, and date and time of collection and date of last ejaculation on the label.
4. Bring specimen directly to the laboratory within one hour of collection (1/2 hour if possible). The specimen must be examined fresh or else sperm may die and no useful information will be obtained.

NOTE: The specimen must be maintained at body temperature during transport. You may do this by keeping it next to your body, in a shirt pocket, for example. Extreme heat or cold may change the test results.

-----  
Please Bring this form with you when you deliver the sample:

Patient Full Name: \_\_\_\_\_

Patient Date of Birth: \_\_\_\_\_

Days of Abstinence: 2 – 7 days? YES NO

Was the entire specimen collected and submitted in sterile container? YES NO

Date and time sample was collected: \_\_\_\_\_

Method of Collection: Ejaculation? YES NO

Did you experience collection or transport problems? YES NO

(If yes, explain: \_\_\_\_\_)

## SPINAL FLUID

Spinal fluids submitted to the Laboratory for analysis require special care in handling. The fluid is obtained by a surgical procedure that involves not only discomfort to the patient, but also a certain amount of risk of complication from the procedure. It is, therefore, necessary for all personnel handling the specimens to be constantly aware of the need for extreme care in preserving the usefulness of the specimen.

### **General Procedure:**

- 1) Test orders are placed in HIS.
- 2) The specimen(s) is brought to the Laboratory immediately following the spinal tap.
- 3) All CSF specimens ordered for culture will have a gram stain done STAT. There is no need to order a separate gram stain.
- 4) See additional information under Cerebrospinal Fluid.

## **SPINAL FLUID - PEDIATRIC**

All pediatric cerebrospinal fluid (CSF) specimens are cultured routinely whether requested or not. If a specimen of less than 0.5 ml of CSF is obtained, the physician is consulted concerning his/her testing priorities.

## SPUTUM CYTOLOGY SPECIMEN - INSTRUCTIONS FOR PATIENT

Thank you for choosing Brattleboro Memorial Hospital for your laboratory test.

You can help us to do the best work possible for you by following these instructions when collecting your **SPUTUM CYTOLOGY** specimen.

1. You will receive one specimen container for each Sputum Cytology test your doctor has ordered.
2. The specimen should be collected on weekdays only.
3. Just after waking in the morning, rinse your mouth with water.
4. Cough forcefully and vigorously to raise a deep coughed specimen from the lungs..
5. Expectorate the deep coughed specimen into the specimen container (saliva is unsatisfactory.)
6. Label the container with your name, date collected, your date of birth, your doctor's name and place it in the refrigerator.
7. Take the specimen to the laboratory on the day of collection as soon as possible, preferably before 11:00 am.

If you have any questions regarding the collection of your specimen, please call the Laboratory at (802) 257-8311 between the hours of 8:00AM and 3:00 PM.

Thank you very much for your close attention to this procedure.



## **SPUTUM SPECIMEN – INSTRUCTIONS FOR PHYSICIAN OFFICE**

1. The patient should be instructed to expectorate deep from the lungs into a sterile specimen container with a tight-fitting lid. Saliva is unsatisfactory as specimen material. Patients should avoid as much as possible adding saliva to the sputum specimen.
2. 5-10 mL of a single early morning specimen is recommended. Small volumes can be accepted, even for mycobacterial culture. A few drops of purulent material can be accepted for routine culture / gram stain. A minimum of 2 mL is needed for mycobacterial culture. Other volume exceptions should be referred to the Microbiology Supervisor.
3. All expectorated specimens will be evaluated microscopically for the presence of saliva. Unacceptable specimens will be rejected and a new specimen will be required.

## STOOL TRANSPORT GUIDE

Test Type	Preferred Transport	Comments
O & P	PVA (black cap)	Can include Cryptosporidium exam if ordered and Cyclospora
Giardia Ag	Formalin only (Yellow cap)	See Notes below for extended comments
WBC, stool	Formalin only (Yellow cap)	
CDIF (Clostridioides difficile)	Unpreserved stool. (Only a liquid stool) in a sterile cup. See diagram on following page	Submit in sterile / clean container (i.e., urine cup)
Stool Culture	Cary Blair Transport (green cap)	No swabs if Campylobacter ordered. Must indicate special request for pathogenic E. coli. Giardia can also be done on Cary Blair.

### **Antigen testing for Giardia lamblia**

A rapid antigen test for Giardia uses an enzyme-immunoassay technique to detect the presence of Giardia antigen in stool specimens. Giardia is the most frequently seen parasite. It is found in over 98% of the positive ova and parasite testing performed at BMH. Therefore testing only for Giardia is justified in most cases. It is recommended:

1. Initial screening for Giardia antigen be ordered
2. If the antigen test is negative and symptoms persist, a second sample be submitted for repeat antigen testing and a complete ova and parasite analysis.
3. If a patient has traveled out of the country a complete ova and parasite analysis should be ordered in addition to the antigen screen. The patient history of travel should be noted on the lab order form.

For all Ova and parasite testing, either antigen screens or an Ova and parasite analysis, it is important that multiple specimens be spaced out at least 1-2 days due to the intermittent nature of the shedding by the parasites.

Collection of specimens should be prior to patient's taking antacids, kaolin, oily laxatives, antidiarrheal medication, mineral oils, barium or bismuth used in radiology procedures, and broad spectrum antimicrobials.

If the patient has taken any of the above treatments, 5-10 days should elapse before collection of stool specimens .

# C. diff Ticket to Ride, Implemented September 2018

## C-DIFF TICKET TO LAB

- Please attach this trip ticket to *all* C - diff stool specimens sent to lab.
- Do not send specimen to lab unless *all* boxes are verified and checked.

YES, there are more than 3 watery stools in the past 24 hours.

**STOOL MUST BE TYPE 7 SEE BELOW**








YES, in the past two weeks during this admission, there haven't been *any* positive C - diff tests.

YES, the patient has not had stool softeners, laxatives, or bowel prep in the past 48 hours.

**IF ALL BOXES ARE NOT CHECKED CANCEL THE TEST IN EPIC**

NURSE/RESIDENT: \_\_\_\_\_ EXT: \_\_\_\_\_ DATE: \_\_\_\_\_

### Bristol Stool Scale

Type 1		Separate hard lumps, like nuts, hard to pass.
Type 2		Sausage-shaped but lumpy.
Type 3		Like a sausage but with cracks on its surface.
Type 4		Like a sausage or snake, smooth and soft.
Type 5		Soft blobs with clean-cut edges (passed easily).
Type 6		Fluffy pieces with ragged edges, a mushy stool.
Type 7		Watery, no solid pieces. Entirely liquid.

Patient Label



### Bristol Stool Chart

Type 1		Separate hard lumps, like nuts (hard to pass)
Type 2		Sausage-shaped but lumpy
Type 3		Like a sausage but with cracks on its surface
Type 4		Like a sausage or snake, smooth and soft
Type 5		Soft blobs with clean-cut edges (passed easily)
Type 6		Fluffy pieces with ragged edges, a mushy stool
Type 7		Watery, no solid pieces. Entirely liquid

## **UNUSUAL OR ONE-TIME SPECIMENS**

Specimens collected by surgical or other invasive techniques or collected by methods that result in unusual patient discomfort are considered one-time; that is, repeating the procedure would be of such difficulty as to be detrimental to patient care.

In order to safeguard these “special specimens”, the Laboratory saves them under refrigeration, or by other appropriate means, for at least 24-hours. In this manner, errors of omission in requesting or in performing tests are minimized.

## URINE COLLECTION (2 Pages)

BMH provides 24-hour urine collection containers. Nursing units can call the Laboratory at ext. 7401 for a collection container.

- Whenever a urine preservative must be used, advise the patient in advance of any potential hazards that might arise from inadvertent spillage, particularly if the preservative is acid. The container must be kept out of reach of children.
- Instruct the patient to discard the first morning specimen and to record the date and time of voiding on the urine container.
- The patient should collect all subsequent urine voided for the remainder of the 24 hours.
- Collect the first morning specimen on day two at the same time as recorded on day one. Record the date and time on the urine container.
- Keep the urine refrigerated or chilled for the full duration of the collection period.
- Inpatient population – bring the urine container to the Laboratory as soon as possible following the last collected specimen and place it in the Laboratory hall refrigerator in the appropriate section.
- The urine container is labeled as follows:
  - a. Patient's full name and DOB
  - b. Date & Time collection began
  - c. Date & Time collection completed
  - d. Patient's room number (Inpatients)
  - e. Name of test to be performed
  - f. Physician / Health Care Provider
  - g. Medical record number (Inpatients).
  - h. Height / weight for creatinine clearances.

**RANDOM MIDSTREAM COLLECTIONS:** For routine analysis and urine culture, instruct the patient to start urinating directly into the toilet; stop, position the container and collect the remaining urine, and to not touch the container to the genital area.

**NOTE:** See Patient Instructions for collecting midstream Urine Specimens

The specimen should be capped, labeled and delivered to the Laboratory as soon as possible (inpatient) or refrigerated until courier pickup time.

## URINE COLLECTION (24-HOUR) – PATIENT INSTRUCTIONS

Your doctor has requested a test that requires a 24-hour URINE COLLECTION.

Please follow these instructions for the collection.

1. Do not void directly into the container; use the urine cup provided to add to the large 24-hour collection container.
2. Consult your physician before discontinuing any medications you are currently taking.
3. Avoid alcoholic beverages and limit caffeine for at least 24 hours prior to the start of the collection and during the collection.
4. Do not exceed your normal intake of liquids during the day before and during the collection period, unless instructed by your doctor to do so.
5. Some collection containers may have preservatives in them containing strong acid. Do not dispose of the preservative. Keep the container out of the reach of children and **avoid contact with the preservative. In case of skin contact, rinse exposed area well with cool water.**
6. The 24-hour collection period begins when you wake up in the morning and empty your bladder. Do not save the first morning specimen. Do write the “start” date and time on the 24-hour collection container. Collect all urine, day and night, for the next 24-hour period. The final collection will be at the same time on the next morning. **Write the “end” date and time on the 24-hour collection container.**
7. Keep the urine collection container cool at all times by refrigerating or keeping it on ice.
8. Return the collection container as soon as possible to the BMH Laboratory Reception Desk. Be sure the start and end date/time are recorded on the label on the container.
9. If your doctor orders a CREATININE OR UREA CLEARANCE: A blood specimen must be drawn within 24 hours of the urine collection. Your HEIGHT and WEIGHT must be recorded on the 24-hour collection container before returning the container to the Laboratory.

If you have any questions, please call the Laboratory at 257-8311. Thank you.

**URINE COLLECTION (24-HOUR) / PRESERVATION (2 Pages)**

<b>TEST ORDERED</b>	<b>PRESERVATIVE</b>
Aldosterone	Refrigeration
Amino Acids (Screen or Panel)	Refrigeration
Aminolevulinic Acid	Refrigeration
Arsenic, Total	Refrigeration
Calcium	Refrigeration
Catecholamines, Fractionated	Refrigeration
Chloride	Refrigeration
Chromium	Refrigeration
Citrate	Refrigeration
Copper	Refrigeration
Cortisol, Free	Refrigeration
Creatinine	Refrigeration
Creatinine Clearance	Refrigeration
Cystine	Refrigeration
Glucose, Quant.	Refrigeration
5-HIAA	Refrigeration
Heavy Metals	Refrigeration
Histamine	Refrigeration
Homovanillic Acid	Refrigeration
17-Hydroxycorticosteroids	Refrigeration
Hydroxyproline, Free or Total	Refrigeration
Iodine, Free	Refrigeration
17-Ketosteroids, Fractionated	Refrigeration
Kidney Stone Formation	Refrigeration
Lead	Refrigeration
Magnesium	Refrigeration
Manganese	Refrigeration
Metanephrines, Fractionated	Refrigeration
Mercury	Refrigeration

<b>TEST ORDERED</b>	<b>PRESERVATIVE</b>
1 or 3-Methylhistidine	Refrigeration
Microalbumin	Refrigeration
Nitrogen	Refrigeration
Oxalate	Refrigeration
Phenylalanine	Refrigeration
Phosphorus	Refrigeration
Porphobilinogen Quant.	Refrigeration
Porphyrins, Fractionated	Refrigeration
Potassium	Refrigeration
Protein, Total	Refrigeration
Protein Electrophoresis	Refrigeration
Sodium	Refrigeration
Thallium	Refrigeration
Urea	Refrigeration
Urea Clearance	Refrigeration
Uric Acid	Refrigeration
Vanillylmandelic Acid (VMA)	Refrigeration



## URINE SPECIMEN (MIDSTREAM) – INSTRUCTIONS FOR PATIENTS

### A. Female, Circumcised and Uncircumcised Males

- 1) Wash your hands thoroughly with soap.
- 2) Open sterile package containing the plastic cup.
- 3) Start urinating into the toilet and then catch the remainder of the urine in the plastic cup. Place the cap on the container without touching its underside.

**CAUTION: Be sure the cap is firmly in place before you leave.**

## **URINE CULTURE - PRESERVATIVE TUBE INSTRUCTIONS (UCT)**

1. Have patient urinate into a sterile container using instructions for clean catch urine.
2. Immediately transfer 5 to 10 mls of urine into the tube and recap tightly. (Adding less than 5 mls of urine may result in loss of viability of bacteria)
3. Label with patient's name, date and time of collection.
4. Specimen is ready to be transported or stored for up to 48 hours at room temperature.

**This specimen is not acceptable for urinalysis, only acceptable for urine culture.**

## VENIPUNCTURE PROCEDURE – ADULT (3 Pages)

### **Purpose**

To provide instructions for the proper collection of patient blood specimens.

### **Supplies Required**

- Collection vacutainer tubes
- Latex-free Tourniquet
- Alcohol pad, chlorhexidine swabs (for blood cultures; Providine for patients < 2 months)
- Disposable vacutainer holder
- Lancet – pediatric collection (Tenderfoot)
- Microtainer – pediatric collection
- Gloves
- Butterfly (winged infusion set)
- Safety needles
- Antibacterial hand disinfectant

### **Procedure**

1. Check inpatient rooms for possible posted isolation or specific blood draw restrictions before and after entering patient room when performing inpatient draws.
2. Wash your hands and wear gloves when working with a patient. Wash hands and change gloves between each patient. Antibacterial hand disinfectant may be used when handwashing is unavailable.
3. The phlebotomist will use two patient identifiers before drawing blood.
  - a) **Inpatient:** Verify the patient's name and Medical Record # on ID bracelet, with draw list and/or labels. Proper identification of patient is crucial. Conscious patients are asked for their full name and date of birth. Compare this to the doctor's orders. While it is always preferred that patients state their names, if a patient is unable to do so, both identifiers can come from the patient's wristband. Having a nurse or family member identify the patient is another option.
  - b) **Outpatient:** Ask the patient a direct question, "What is your name, sir/madam?" and "What is your date of birth?" Compare the information stated by the patient with the information on the computer labels or with the requisition slip. Family member or caregiver may also verify the patient's identity.
4. Position the patient's arm straight from the shoulder to wrist, resting on the arm of the chair.
5. Verify correct tubes are selected and supplies are within reach.
6. Ask the patient to close his/her hand gently. Avoid a clenched fist or vigorous pumping of the hand.

**Note:** Areas to avoid when selecting a site:

- a) Extensively scarred area
- b) Hematoma area
- c) Side where a mastectomy was performed
- d) Cannula, fistula vascular graft arm

7. Palpate and trace the path of veins with your index finger. Veins lack resilience, feel cord-like and roll easily. If superficial veins are not apparent, massage the arm from wrist to elbow, tap the vein site or apply a warm cloth to the area.
8. Clean the area with alcohol in a circular motion from the center to the periphery. To avoid discomfort to the patient and hemolysis of specimen, allow area to dry prior to venipuncture. If the site must be palpated again, clean using a new alcohol prep.
9. Wrap the tourniquet around the arm 3-4 inches above the venipuncture site on skin or around "light" clothing. Never leave the tourniquet on for an extended period of time.
10. Examine the needle to ensure that it is free of hooks and its opening is clear of any particles that may obstruct the flow of blood.
11. Grasp the patient's arm firmly. Anchor the vein with your thumb 1-2 inches below the venipuncture site.
12. Tap all tubes with additives to ensure that the additive is dislodged from the stopper.
13. Line up the needle (bevel up) with the vein. Perform the venipuncture with a smooth entry. Once blood flow is established, instruct the patient to open his/her hand and remove the tourniquet.
14. Fill each tube until the vacuum is exhausted. Gently invert tubes with additives 5-10 times according to manufacturer guidelines. To avoid hemolysis, do not mix vigorously.
15. If the specimen cannot be obtained, follow troubleshooting tips (i.e., change position of needle, try another tube, loosen the tourniquet). If this does not work, select another site. **DO NOT ATTEMPT MORE THAN TWO SITES. If unable to obtain an adequate specimen, two more attempts by 1 or 2 phlebotomist(s) may be allowed, but no more than a total of four attempts.**

**NOTE: If after four unsuccessful attempts, Lab Supervisor and Nursing Unit must be notified. If an outpatient, notify the patient's healthcare provider. This notification must be documented.**

16. Apply slight pressure to the site with gauze and smoothly withdraw the needle. Instruct the patient to apply pressure or bend his/her arm up until bleeding stops. Once bleeding has stopped, apply a bandage or tape gauze to the area. Instruct the patient to leave it on for at least 15 minutes. If bleeding continues, hold direct pressure on the site until bleeding stops and alert the physician. "Coban" bandage may be used for patients on Coumadin or aspirin therapy.
17. Needles and vacutainer holder must be disposed of immediately in an approved sharps container. Recapping needles and vacutainer holder reuse is prohibited.
18. Label tubes at the bedside or next to OP immediately after collection with patient's full name, date and time of collection, and your initials/tech code.

## **USE OF BUTTERFLY NEEDLES**

A winged collection set (butterfly) is not for routine use. They are primarily used for blood culture collections. They may also be used on veins of infants and small children, or on difficult or hand veins of adults. Butterfly usage should be considered a last resort.

## **VENIPUNCTURE - PEDIATRIC (4 Pages)**

Venipuncture should be avoided in children under the age of two (2) years. Venipuncture is performed only if the specimen volume required exceeds the volume obtainable through capillary puncture.

If the child is under 2 years of age, the site should be limited to superficial veins and assistance provided to restrain the child during collection. Take special care to secure the arm to prevent injury caused by unexpected movement.

Definition of skin puncture: a mixture of blood from arterioles, venules and capillaries containing interstitial and intercellular fluids.

Identification of pediatric patients and cleaning of the site is the same as for adults.

Skin puncture site newborn: Use the plantar surface medial to a line drawn posteriorly from the middle of the great toe to heel, or lateral to a line drawn posteriorly from between the fourth and fifth toes to heel. See Figure 3. Warm the foot prior to skin puncture with a warm cloth or heel warmer. The curvature of the heel should be avoided.

Skin puncture site three to 12 months of age: use the plantar surface of the big toe. Warm the infant's foot with a warm cloth or heel warmer prior to skin puncture.

Previous puncture sites should be avoided.

Skin puncture site one year to adult: use the palmer surface of the middle finger. See figure 2.

### **PREPARATION & ID:**

1. Phlebotomist Preparation:
  - Requisition form must contain the same information as required for venipuncture.
  - Phlebotomist should carefully examine the information on the requisition form.
2. Patient Identification and Preparation:
  - Identify the patient
    - In the nursery, check baby's ID band for name and MR#.
    - On OP's, Verbal ID by parent/guardian to include name and DOB.
    - 2 independent identifiers are required (room / bed # disallowed).
3. Give parents choice of staying with the child or leaving the room.
4. Approaching pediatric patients can be difficult. Be friendly and confident. Explain necessity of remaining still during procedure. Parental consent must be obtained if a restraint is used.

### **PROCEDURE:**

1. Hold the foot or hand firmly to prevent movement during collection.

2. After the site has been chosen and prepared, puncture the skin. Wipe off the first drop of blood with gauze. Gently apply pressure to surrounding area during collection.  
**Note:** To avoid hemolysis, do not use strong pressure.

Site Selection:

- Primary danger in skin puncture is accidental contact with bone followed by infection (osteomyelitis). Because the tip and sides of fingers contain only about half the tissue mass of the central area, the possibility of bone injury is increased.
  - Primary Sites are:
    - Heel
    - Distal segments of the 3<sup>rd</sup> & 4<sup>th</sup> fingers
  - Non-recommended sites:
    - Plantar surface of large toe
    - Ear lobes
    - Thumb has possible calluses
    - Index finger has increase in nerve endings
    - 5<sup>th</sup> finger has decreased tissue
  - Punctures should **NEVER** be made through previous puncture sites (may introduce bacteria into the puncture).
  - Warming the area:
    - Use a moistened towel with warm water (40°C) or
    - By activating a commercial heel warmer and covering the site for 3 to 5 minutes.
3. Performing the Puncture:
- The heel or finger should be well supported and held firmly.
  - Massaging the area before the puncture may increase blood flow to the area.
  - The heel is held between the thumb and index finger of the nondominant hand, with the index finger held over the arch and the thumb below the heel.
  - The finger is held between the thumb and the index finger with the palmar surface facing up.
  - Automatic devices should be placed firmly on the puncture site.
  - The blade of the lancet should be aligned to cut across the groove of the finger or heel print. This aids in the formation of a rounded drop as the blood will have a tendency to run into the grooves.
  - Place used lancet in an appropriate sharps container.
  - Once collection is complete, wipe the area with gauze and apply pressure to stop bleeding. A bandage is not necessary after bleeding has stopped.
  - Label specimens and note on the orders: capillary specimen.
4. Specimen Collection:
- Wipe the first drop of blood away using a sterile gauze.
  - Blood should be freely flowing from the puncture site as a result of firm pressure.
  - Collection containers fill by capillary action.
  - **DO NOT** touch the puncture site with the collection device.
  - Fingers are positioned slightly downward but with the palmar surface facing downward during the collection procedure.
  - To prevent the introduction of air bubbles, capillary tubes and micropipets are held horizontally while being filled.

- Microcollection tubes are slanted downward during the collection procedure and blood is allowed to run through the capillary collection scoop and down the side of the tube.
- Once collection is complete, wipe the area with gauze and apply pressure to stop bleeding. A bandage is not necessary after bleeding has stopped.
- Label as a capillary specimen

**NOTES:**

1. Patient questions concerning the need or significance of tests ordered should be directed to the physicians.
2. Steps to take for prevention of hematoma:
  - Ensure the needle fully penetrates the uppermost wall of the vein.
  - Remove the tourniquet before removing the needle.
  - Apply slight pressure with gauze when bandaging.
3. Steps to take for prevention of hemolysis:
  - Mix specimens with additives gently but thoroughly.
  - Avoid collecting from a site with a hematoma present.
  - Avoid using a needle that is too small (use butterfly needles only when absolutely necessary).
  - Make sure the needle is fitted securely to the vacutainer holder.
  - Allow alcohol to dry before venipuncture is performed.
4. The recommended order of draw when collecting multiple tubes is:
  - Pediatric blood culture tubes.
  - Red top for non-additive or serum tubes.
  - Light blue top for citrate tubes.
  - Gel-separation and clot activator tubes.
  - Green top for tubes containing heparin.
  - Lavender top for EDTA tubes.
  - Gray Top for lactic acid.
  - Other additive tube depends on the manufacturer. Be sure to consult the manufacturer's package insert.
5. If coagulation testing (PT/PTT/Fibrinogen) is the only test to be collected and you are using a butterfly, draw blood into a red top tube until blood enters the tube to eliminate the air, and then can discard the tube. There is no need to fill the tube. This process eliminates air and ensures the tube for testing has the correct volume. If at all possible, avoid using a butterfly needle.
6. Collection of blood for coagulation testing through IV lines that have been previously flushed with heparin should be avoided, if possible. If the blood must be drawn through an indwelling catheter, possible heparin contamination and specimen dilution should be considered. When obtaining specimens from indwelling lines that may contain heparin, the line should be flushed with 5-10 mL of saline and the first 5 mL of blood or 6-times the line volume (dead space volume of the catheter) be drawn off and discarded before the coagulation tube is filled. Laboratory Staff cannot draw via IVs or catheters.



## TECHNIQUE FOR PUNCTURING THE SKIN

FIGURE 2  
TECHNIQUE FOR PUNCTURING THE SKIN

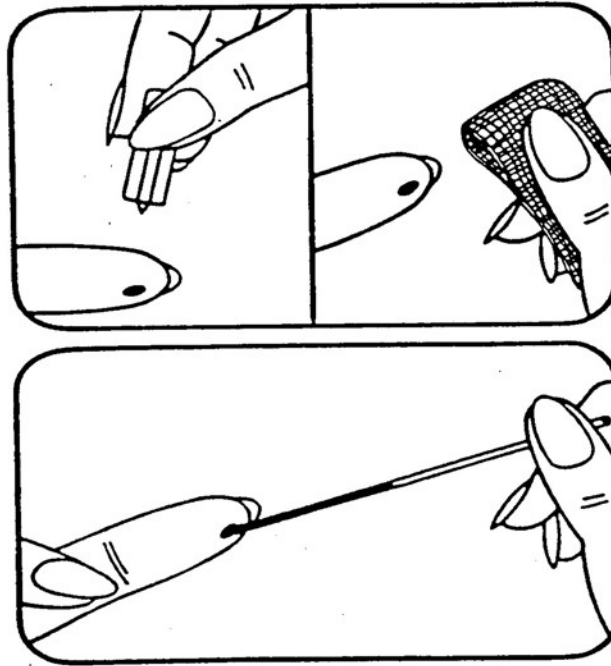
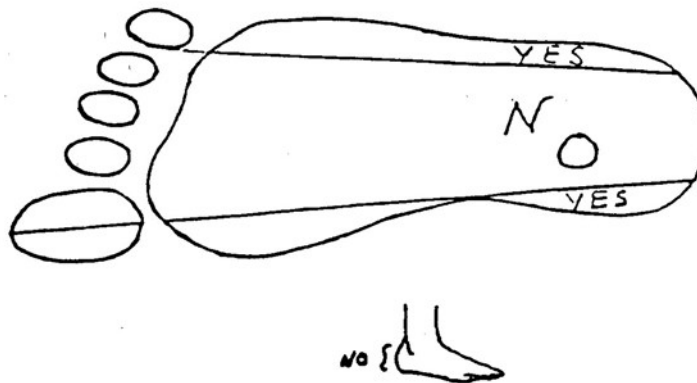


FIGURE 3  
Recommendation for Heel Skin Puncture in Newborn Infants



1. Perform punctures on the most medial or most lateral portion of the plantar surface (outside of the lines on the diagram).
2. Puncture no deeper than 2.4 mm.
3. Do not perform punctures on the posterior curvature of the heel.
4. Do not puncture through previous sites which may be infected.

## **PATIENT CARE**

The Laboratory Staff may provide the following patient care:

### **Fainting:**

5. Lower the patient's head or transfer to bed.
6. Loosen tight clothing.
7. Apply cold compresses to forehead and back of neck.
8. Call 111 and ask for a Rapid Response.

### **Nausea:**

4. Make patient as comfortable as possible.
5. Instruct patient to breathe deeply and slowly.
6. Apply a cold compress to forehead.

### **Vomiting:**

3. Provide a basin and tissues.
4. Give patient water to rinse out mouth.

### **Convulsions:**

4. Prevent patient from injuring him/herself.
5. **Do not** restrain patient.
6. Notify the physician for immediate assistance.

## WET MOUNT

### Microscopy

**Steps** — A sample of the patient's vaginal discharge is obtained with a cotton swab, smeared onto a slide, and evaluated under a microscope with both saline and potassium hydroxide in the steps below. Subsequent treatment is determined by the findings of the microscopic evaluation.

- **Saline wet mount** – Vaginal discharge is generally sampled with a plastic or wood vaginal/cervical scraper or a cotton-tipped swab. The sample of vaginal discharge is mixed with one to two drops of 0.9 percent normal saline solution at room temperature on a glass slide. Cover slips are then placed on the slides, which are examined under a microscope at low and high power. Microscopy should be performed within 10 to 20 minutes of obtaining the sample to reduce the possibility of loss of motility of any trichomonads. (From UpToDate April 4/8/2019)

**SPECIAL COLLECTION INFORMATION FOR SPECIMENS SENT TO A  
REFERENCE LAB (2 pages)**

**BREAST CANCER PROFILE**

**SPECIMEN REQUIREMENT:** Paraffin sections of tissue positive for tumor.  
**ORDERABLE TESTS:** ER, PR, Her-2/NEU (Tests may be ordered individually)

**CHROMOSOME ANALYSIS**

**SPECIMEN REQUIREMENT:** 3 cm<sup>3</sup> products of conception, skin or fascia.  
1 cm<sup>3</sup> of placenta (including chorionic villi).  
Patient history required.

**COLLECTION REQUIREMENT:** Sterile container containing sterile saline. No formalin.  
Avoid contamination.

**LIMITATIONS:** If specimen does not consist of viable products of  
conception, there may be no cell growth.

**STORAGE REQUIREMENT:** Refrigerate (Do not freeze).

## CYTOLOGY SPECIMENS (NON – GYN)

<b>Body Fluid Cytology</b>	<b>Refrigerate until ready to send</b>
<b>Pleural, Pericardial, Peritoneal</b>	<p>All specimens should be submitted in sterile containers. If the clinician needs to add an anticoagulant, heparin or EDTA may be used. There is no specific volume requirement.</p> <p>Do not send glass container; instead pour specimen into 2 sterile urine containers and write the original volume of the sample on the requisition.</p>
<b>Bronchial Washing Cytology</b>	<p>25 mL is requested – especially if Microbiological culture is also needed.</p> <p>Add an equal volume of CytoRich Red after removing a portion for culture (if needed).</p>
<b>Bronchial Brushing</b>	Brushes should be submitted in CytoRich Red.
<b>Cerebrospinal Fluid</b>	<p>CSF should be sampled into 3 or 4 sterile, sequentially numbered tubes at time of collection. All samples should be sent to the laboratory as soon as possible after collection. CSF should be refrigerated until they are sent. If the CSF sample for cytology cannot be sent to the referral laboratory the same day, then add an equal volume of CytoRich Blue.</p>
<b>Gastrointestinal Brushings</b>	Brushes should be submitted in CytoRich Red
<b>Nipple Discharge</b>	<p>The slides are prepared by the clinician and should be fixed in alcohol or spray fixed immediately. Slides will be stained at BMH. It is not necessary to refer slides to DHMC- Keene.</p>
<b>Sputum Cytology</b>	<p>Send fresh if it is going to be sent the same day as collected. If the courier has already left for the day, or on weekends, add an equal volume of CytoRich Red and store at room temperature.</p>
<b>Urine</b>	<p>Send fresh if it is going to be sent the same day as collected. If the courier has already left for the day, or it is a weekend then add an equal volume of CytoLyte to the urine container. Some urine can be poured off for a final urine volume of 25 mL because the CytoLyte is prepared in 25 mL aliquots. If the urine is diluted in alcohol, there is no need to add CytoLyte. Alcohol solution should be noted on the container.</p>

## CYTOLOGY SPECIMENS (GYN)

### *THIN PREP CYTOLOGY MATERIALS*

Sampling devices and PreservCyt Thin Prep specimen collection vials are obtained from the main laboratory.

## FLOW CYTOMETRY, LYMPH NODE

<b><u>SPECIMEN REQUIREMENT:</u></b>	Lymph node biopsy.
<b><u>COLLECTION REQUIREMENT:</u></b>	Fresh
<b><u>LIMITATIONS:</u></b>	<b>Adequacy of sample.</b>
<b><u>STORAGE REQUIREMENTS:</u></b>	<b>Deliver immediately to Main Laboratory.</b>

## KIDNEY BIOPSY for TRANSPLANT

<b><u>SPECIMEN REQUIREMENT:</u></b>	Tissue. Call Laboratory at 257-8311 24 hours before collecting specimen so lab can order correct fixative from reference lab if necessary. Patient history required. The ordering physician will need to submit a kidney biopsy to the on-call Pathologist for an immediate consultation.
<b><u>STORAGE REQUIREMENT:</u></b>	<b>Must immediately</b> be examined in the Pathology Department in sterile saline.

## SKIN BIOPSY, IMMUNOFLUORESCENCE

<b><u>SPECIMEN REQUIREMENT:</u></b>	Tissue Biopsy: Specimen must be received in the laboratory within 24 hours. Notify Anatomic Pathology before collecting specimen (preferably a few days' notice if possible).
<b><u>COLLECTION REQUIREMENT:</u></b>	Pre-filled container of transport media (obtain from Lab, 257-8311).
<b><u>STORAGE REQUIREMENTS:</u></b>	Refrigerate.

## STONE ANALYSIS

<b><u>SPECIMEN REQUIREMENT:</u></b>	Unfixed specimen on saline-soaked gauze Requisition must state site of origin.
<b><u>COLLECTION REQUIREMENT:</u></b>	Sterile plastic container.

## URINE CYTOLOGY SPECIMEN – INSTRUCTIONS FOR PATIENT

Thank you for choosing Brattleboro Memorial Hospital for your laboratory test.

You can help us to do the best work possible for you by following these instructions when collecting your URINE CYTOLOGY specimen.

1. You will receive one sterile specimen container containing for each Urine Cytology test your doctor has ordered.
2. Urine samples should be collected on weekdays only.
3. Collect your specimen from the second morning voided urine into the sterile specimen container provided.
4. If necessary, you may drink one glass of water every 30 minutes for three hours until you are able to collect a specimen.
5. Add the specimen you have collected to the specimen container.
6. Label the container with your name, date collected, your date of birth, your doctor's name and place it in the refrigerator.
7. Take the specimen to the laboratory on the day of collection as soon as possible, preferably before 11:00 am.

Please consult your doctor if you are unable to void after taking the steps described above.

If you have any questions regarding the collection of your specimen, please call the Main Laboratory at (802) 237-8311, between the hours of 8:00AM and 4:30PM.

Thank you very much for your close attention to this procedure.