

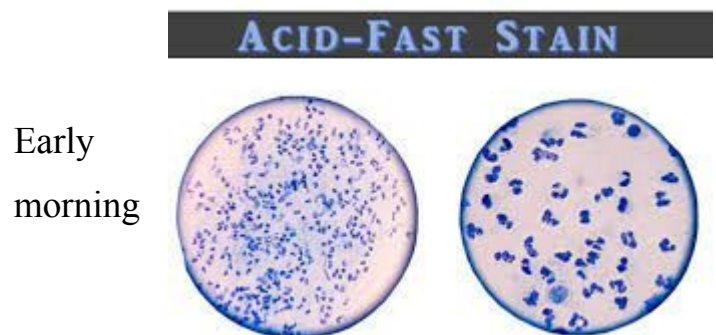
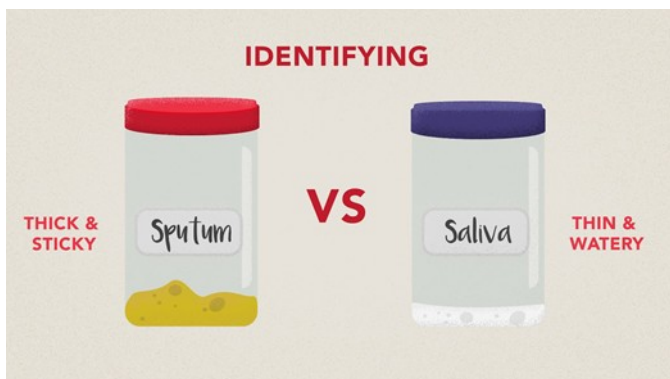
LAB 5 : Specimen Collection & Processing

➤ Types of Medical Specimens:

- Pus, ulcer & skin specimens (hair-nail-wounds.....etc.).
- Eyes, nose & mouth specimens.
- Upper & lower respiratory tract specimens.
- Urogenital specimens (semen-urethral discharge- vaginal discharge...etc.)
- Faecal specimens.
- Urine specimens.
- Blood specimens.
- Effusion specimens.
- Cerebrospinal fluid (CSF) Specimens.

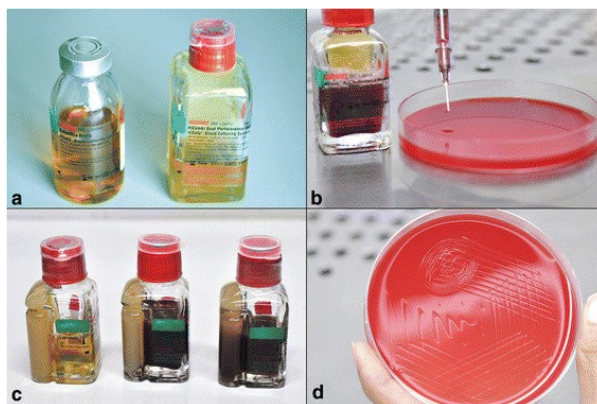
➤ Collection Times

1. The optimal times for specimen collection must be based upon both the type of infectious disease process and the ability of the laboratory to expertly process samples. Laboratories are usually better staffed during **daytime** hours to receive specimens.
2. Pathogens in highest concentration in first morning collections will be diluted by added secretions. There is a high likelihood that samples stored after collection may become overgrown with contaminants.
3. The first early morning sputum and urine samples are optimal for recovery of **acid-fast bacteria, fungi, and other pathogens. Samples collected at other times are acceptable.**



secretions are more concentrated and more likely to contain large numbers of the etiologic agent.

4. The timing of **blood cultures*** should be determined by the clinical condition of the patient. Physicians should always indicate the collection schedule. Except in acute cases of septicemia, blood cultures should not be drawn more frequently than 1/2 h apart. A total of three cultures per 24 h is usually sufficient to diagnose most cases of septicemia.



➤ Specimen Transport

Microorganisms are living things—rapidly they **grow**, they **reproduce**, they **die**. **Transport media** are designed to prevent or slow all **three processes**. Incomplete or misleading laboratory data may result if any of the three occur before the specimen can be cultured in the laboratory. Please hurry; the work can't be started until the specimen arrives

Stuart's medium (1954)

1. Originally formulated for transport of *Neisseria gonorrhoeae*.
2. Used charcoal-impregnated swabs which caused difficulty in Gram stain interpretation. *
3. nonnutritive medium



Amies medium (1965)

1. Modified Stuart's medium.
2. Replaced glycerophosphate with a balanced salt solution.
4. Better transport system for most specimens.



Cary & Blair medium (1964)

1. Similar to Stuart's but modified for fecal specimens.
2. pH increased from 7.4 to 8.4.
3. Removed charcoal from formula.
4. Good for stool specimens.
5. Recommended for fecal specimens suspicious for *Campylobacter* sp. and for other enteric pathogens.



Buffered glycerol saline medium

1. Designed for stool specimens only.
2. Good for mailing fecal specimens.
3. High pH to favor fecal pathogens.
4. Not for transport of fecal specimens in which *Campylobacter* sp. is suspected.

➤ Specimen Refrigeration

1. Specimens that **CAN** be refrigerated before inoculation of media:

- **Urines »Respiratory Exudates**
- **Stools/Feces «Wounds**

2. Specimens that **CANNOT** be refrigerated before inoculation of media:

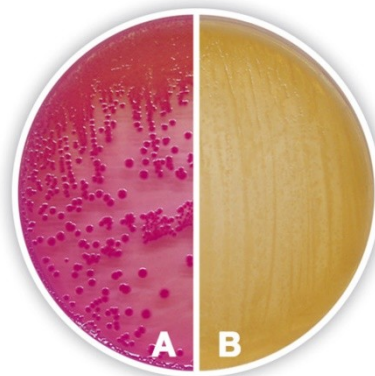
- **Spinal Fluids and Other Body Fluids**
- **Genital/Cervical for Gonococcus Isolation**
- **Blood**

If processing is delayed, spinal fluids should be held at 35°C.

➤ Specimens Processing

Specimen Processing in Bacteriology

Type of Specimen	Media and Conditions	Normal Flora	Common Pathogens	Comments
Throat	Blood agar 35°C aerobic Chocolate agar (for pediatric) 35°C, CO ₂	Alpha and gamma strep Commensal <i>Neisseria</i> <i>Staphylococcus epidermidis</i> Diphtheroids <i>Streptococcus pneumoniae</i> Anaerobes <i>N. meningitidis</i>	Group A, beta-hemolytic streptococci <i>(Haemophilus influenzae)</i> <i>(Corynebacterium diphtheriae)</i> <i>(Bordetella pertussis)</i>	
Respiratory	Blood agar Chocolate agar 35°C, CO ₂ MacConkey (aerobic) Anaerobic media.....	Larynx, trachea, sinus, Sputum: <i>S. epidermidis</i> Non-beta streptococci Diphtheroids Commensal <i>Neisseria</i> Haemophilus Transtrachael and Bronchial: None	Group A, beta streptococci <i>H. influenzae</i> <i>S. aureus</i> Enterobacteriaceae <i>Pseudomonas</i> sp. <i>S. pneumoniae</i>	Gram stain: Sputum All aspirates
Urine Clean catch specimen colony count Catheterized or aspirate	Blood agar MacConkey 35°C, aerobic	None	<i>Escherichia coli</i> and other enterics Enterococci <i>Pseudomonas</i> sp. Staphylococci	Identify all isolates from catheterized specimens



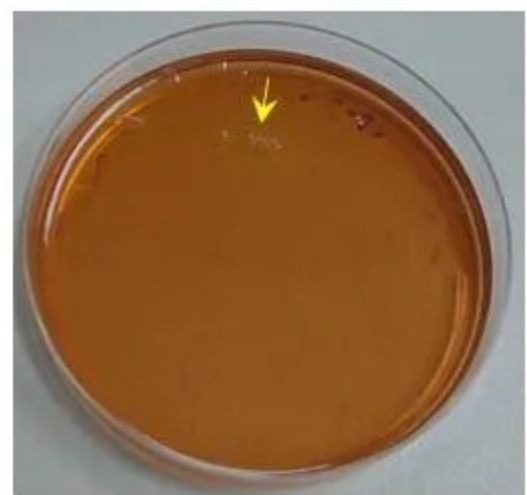
Feces	MacConkey Xylose-Lysine- Desoxycholate Selenite F 35°C, aerobic Selective medium for Campylobacter 42°C, 5% O ₂	Anaerobes Enterobacteriaceae Enterococci	<i>Salmonella</i> sp. <i>Shigella</i> sp. <i>Arizona</i> sp. <i>Yersinia enterocolitica</i> <i>Campylobacter jejuni</i> <i>E. coli</i>	
Genital for GC only	Modified Thayer-Martin (MTM) 35°C, CO ₂	Lactobacilli Diphtheroids Alpha streptococci Enterobacteriaceae Enterococci	<i>Neisseria gonorrhoeae</i>	Gram stain inhibited on MTM
Vaginal, cervical, routine genital	Blood agar Enriched chocolate MTM 35°C, CO ₂ MacConkey - aerobic Anaerobic blood agar	Lactobacilli Diphtheroids Enterococci Enterobacteriaceae Non-beta streptococci Anaerobes <i>G. vaginalis</i>	<i>N. gonorrhoeae</i> <i>Candida albicans</i> <i>Gardnerella vaginalis?</i> Group A and B streptococci <i>Listeria monocytogenes</i>	Culture for <i>G. vaginalis</i> is the prerogative of each labora- tory.
Genital – surgical or aspirates	Blood agar Chocolate agar MTM 35°C, CO ₂ MacConkey - aerobic Anaerobic blood agar	None	Same as above plus anaerobes	Gram stain



Escherichia coli



Salmonella



Shigella

Type of Specimen	Media and Conditions	Normal Flora	Common Pathogens	Comments
Sterile body fluids CSF, joint fluid, pleural fluid, peritoneal fluid, etc.	Blood agar Chocolate agar 35°C, CO ₂ MacConkey agar Thioglycollate 35°C, aerobic	None	Identify all isolates <i>S. pneumoniae</i> <i>N. meningitidis</i> <i>H. influenzae</i> Gram-negative rods	Gram stain
Blood	Submitted in two bottles: one 35°C aerobic and one 35°C anaerobic	None	Any isolate potentially significant	Gram stain
Wound (superficial) includes eye and ear	MacConkey agar 35°C, aerobic Blood agar Chocolate (eye) 35°C, CO ₂	<i>S. epidermidis</i> Diphtheroids Commensal <i>Neisseria</i> Anaerobes Other skin flora	<i>S. aureus</i> Beta-hemolytic streptococci <i>P. aeruginosa</i> <i>H. influenzae</i> Biotype III (<i>H. aegyptius</i>) Enterobacteriaceae	Gram stain
Wound (surgical or aspirate) Tissue specimens	Same as above plus anaerobic blood agar K-V blood agar* Thioglycollate	None	Same as above plus anaerobes Potentially any isolate	Gram stain

*Kanamycin-Vancomycin



beta-hemolysis
Streptococcus pyogenes



alpha hemolysis
Escherichia coli



gamma hemolysis (no hemolysis)
Staphylococcus epidermidis