PROGRAM Standard Operating Procedure – Laboratory Services	
Title: MIC31800 – Urine Culture	Policy Number:
Program Name: Laboratory Services	
Applicable Domain: Lab, DI and Pharmacy Services	
Additional Domain(s):	
Effective Date:	Next Review Date
Issuing Authority: Director of Health Services	Date Approved:
Accreditation Canada Applicable Standard: N/A	

GUIDING PRINCIPLE:
Urine is normally a sterile body fluid. A urinary tract infection is defined by the presence of bacteria in the urinary tract, and by the resulting host inflammatory response (leukocytes or white blood cells in the urine). Significance of growth is dependent upon the number of colony forming units (CFU) present per liter of urine. However, urine is easily contaminated with bacteria from the perineum, prostate, urethra or vagina. Therefore, significance of growth is also partially dependent upon the number of different colony types present.

Bacteria most commonly isolated from patients with uncomplicated UTI's are *Escherichia coli, Klebsiella* species and other *Enterobacteriaceae*, and *Staphylococcus saprophyticus*. Hospitalized patients and those with complicated UTI's are commonly infected with *Escherichia coli*, *Klebsiella* species, *Proteus mirabilis*, other *Enterobacteriaceae*, *Pseudomonas aeruginosa* and *Enterococcus* species.

PURPOSE/RATIONALE:
To determine the presence or absence of bacterial pathogens in urine specimens.

SCOPE/APPLICABILITY:
This procedure applies to Medical Laboratory Technologists (MLTs) processing specimens for urine culture.

SAMPLE INFORMATION:

Type	Urine • Fresh urine collected in sterile container • Fresh urine collected in urine transport tube	
Source	Voided urine (non-sterile)	• Midstream urine (MSU) • Neonatal bagged urine • Indwelling catheter (Foley) urine • Ileal conduit urine
	Aseptically collected urine (sterile)	• Straight, intermittent or "in and out" catheter • Nephrostomy urine • Cystoscopy urine • Suprapubic bladder aspirate
Stability	• Fresh urine collected in sterile container is acceptable for 24 hours, refrigeration necessary • Fresh urine collected in urine transport tube is acceptable for 72 hours, refrigeration not necessary	
Storage Requirements	• Fresh urine - refrigerated • In urine transport tube - room temperature	
Criteria for rejection	1. Fresh urine specimens (orange top) >24 hours old 2. Urine transport tube specimens >72 hours old 3. Unlabeled/mislabeled specimen 4. Specimen container label does not match patient identification on requisition 5. Duplicate specimens obtained with same collection method within 24 hours 6. Leaking specimens 7. Improperly collected, labeled, transported or handled aseptically collected specimens should be processed. Waver of responsibility form SCM40110 needs to be filled out by the responsible nurse	

REAGENTS and/or MEDIA:

- Uri*Select* 4 agar (URI)
- Identification reagents: catalase, oxidase, tube coagulase, etc.

SUPPLIES:

- 1 µL loops
- Wooden sticks
- Glass test tubes
- Sterile pipettes
- Filter paper
- Glass microscope slides

EQUIPMENT

- Biosafety cabinet
- 35° ambient air incubator
- Vitek 2 and supplies

SPECIAL SAFETY PRECAUTIONS:

Containment Level 2 facilities, equipment, and operational practices for work involving infectious or potential infectious materials or cultures.

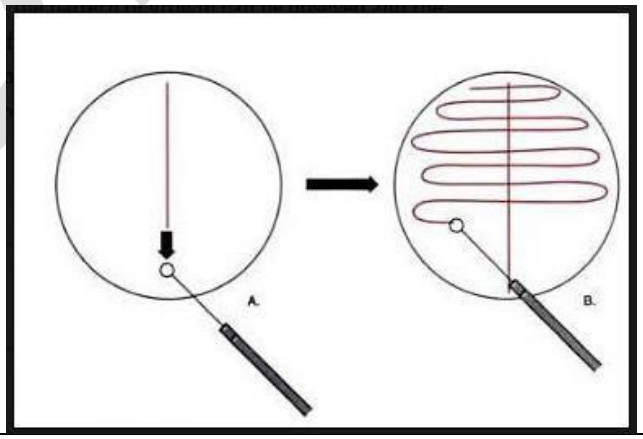
- Ensure that appropriate hang hygiene practices be used.
- Lab gown must be worn when performing activities with potential pathogens.
- Gloves must be worn when direct skin contact with infected materials is unavoidable.
- Eye protection must be used when there is a known or potential risk of exposure of splashes.
- All procedures that may produce aerosols, or involve high concentrations or large volumes should be conducted in a biological safety cabinet (BSC).
- The use of needles, syringes and other sharp objects should be strictly limited.

All patient specimens are assumed to be potentially infectious. Routine Practices must be followed. Since viable micro-organisms are used, all cultures must be handled with appropriate precautions. All equipment in contact with cultures should be decontaminated by appropriate methods.

QUALITY CONTROL:

- Refer to MIC60040 – Culture Media Quality Control procedure
- Refer to Test Manual for reagent quality control procedures

PROCEDURE INSTRUCTIONS:

Step	Action
Processing specimens for urine culture	
1	Hold a 1 µL loop vertically and immerse just below the surface of a well-mixed urine specimen.
2	Deliver a loopful of urine onto the Uri*Select* 4 agar and make a straight line down the center.
3	Streak the urine by making a series of passes at 90° angles through the inoculum: 

	IF	THEN
4	Voided urines (non-sterile): • Midstream urine (MSU) • Neonatal bagged urine • Indwelling catheter (Foley) urine • Ileal conduit urine	• Incubate plate for 18-24 hours at 35° in the O_2 incubator
	Aseptically collected urines (sterile): • Straight, intermittent or "in and out" catheter • Nephrostomy urine • Cystoscopy urine • Suprapubic bladder aspirate	• Incubate plate for 48 hours at 35° in the O_2 incubator

INTERPRETATION OF RESULTS:

- Using a 1 µL loop, 1 colony equals 1 X 10^6 CFU/L
- Determine the colony count and extent of the work-up required for each morphotype on the plate
- Record all observations in the LIS

List of Uropathogens and Non-Uropathogens:

Uropathogens	Potential Uropathogens	Non-uropathogens
Enterobacteriaceae *Pseudomonas aeruginosa* Other GNB *Enterococcus* spp. *Streptococcus pyogenes* *Streptococcus agalactiae* *Aerococcus urinae** *Corynebacterium urealyticum* *Staphylococcus aureus* *Staphylococcus saprophyticus*: (Females, aged 13-55yrs) Yeast spp.	Coagulase negative *Staphylococcus* (Not *Staphylococcus saprophyticus*) **NOTE:** Only considered significant if: ✓ The patient is symptomatic (indicated in clinical history) **AND** ✓ The organism is pure	*Lactobacillus* spp. Diptheroids: (not *C.urealyticum*) Viridans *Streptococci*: (not *A.urinae*) *Bacillus* spp. *Neisseria* spp.

***** Considered a uropathogen only if colony count is 10 times greater than that of all other microbiota

REPORTING INSTRUCTIONS: Non-sterile urine

No. of colonies Colony count	1 isolate (uropathogen or non-uropathogen)	2 isolates (uropathogens or non-uropathogens)	3 or more isolates (uropathogens or non-uropathogens)
≤10 colonies ≤10 X 10^6 CFU/L	Report: **"No Significant Growth"**	Report: **"No Significant Growth"**	Report: **"No Significant Growth"**

No. of colonies Colony count	1 uropathogen	2 uropathogens	3 or more uropathogens
11-99 colonies 11-99 X 10^6 CFU/L	ID and susceptibility	ID and susceptibility on both	Report: **"}CON1" Mixed**
≥100 colonies ≥100 X 10^6 CFU/L	ID and susceptibility	ID and susceptibility on both	Report: **"}CON1" Mixed**

No. of colonies Colony count	1 uropathogen and ≥1 isolates ≤10 (uropathogen or non-pathogen)	2 uropathogens and ≥1 isolates ≤10 (uropathogen or non-pathogen)	≥3 uropathogens and ≥1 isolates ≤10 (uropathogen or non-pathogen)
Uropathogens: >10 Other isolates ≤10	ID and susceptibility on uropathogen Ignore isolate(s) ≤10	ID and susceptibility on uropathogens Ignore isolate(s) ≤10	Report: **"}CON1" Mixed"**

No. of colonies Colony count	1 non-uropathogen	2 non-uropathogens	≥3 non-uropathogens
≤10 colonies ≤10 X 10^6 CFU/L	Report: **"No Significant Growth"**	Report: **"No Significant Growth"**	Report: **"No Significant Growth"**
11-99 colonies 11-99 X 10^6 CFU/L	Report: **"No Significant Growth"**	Report: **"No Significant Growth"**	Report: **"No Significant Growth"**
≥100 colonies ≥100 X 10^6 CFU/L	Report: **"No Significant Growth"**	Report: **"No Significant Growth"**	Report: **"No Significant Growth"**

NOTE: Perform susceptibility testing as per ASTM

REPORTING INSTRUCTIONS: Sterile urine

Colony Count	Any number of morphotypes
Any growth (regardless of number of colony types or count of colonies)	Perform ID and susceptibility testing
No growth after 48 hours incubation	Report: **"}NG2D"**

LIMITATIONS:

1. A mixed culture in an uncomplicated outpatient population likely indicates contamination.
2. For uncomplicated UTI, culture is usually not indicated.
3. False-negative results may be due to interfering substances, diluted urine, low urine pH and subjective interpretation of the criteria for further workup of the culture.

CROSS-REFERENCES:

- MIC60040 Culture Media Quality Control

REFERENCES:

1. Leber, A. (2016). *Clinical microbiology procedures handbook.* (4th ed.) Washington, D.C.: ASM Press
2. Jorgensen J.H., Pfaller M.A., Carroll K.C., Funke G., Landry M.L., Richter S.S., Warnock D.W. (2015). *Manual of Clinical Microbiology, 11th edition*. Washington, D.C: ASM Press
3. BioRad Laboratories. (November 2013). *UriSelect 4* package insert

APPROVAL:

_____

Date

_____

REVISION HISTORY:

REVISION	DATE	Description of Change	REQUESTED BY
1.0	23 Dec 16	Initial Release	L. Steven
2.0	30 Nov 18	Updated to include new Vitek 2 instrument	L. Steven
3.0	31 Dec 20	Procedure reviewed and added to NTHSSA policy template	L. Steven