PROGRAM Standard Operating Procedure – Laboratory Services	
Title: MIC33400 – Genital Culture-IUD	Policy Number:
Program Name: Laboratory Services	
Applicable Domain: Lab, DI and Pharmacy Services	
Additional Domain(s):	
Effective Date:	Next Review Date:
Issuing Authority: Director, Health Services	Date Approved:
Accreditation Canada Applicable Standard: N/A	

GUIDING PRINCIPLE:
Genital colonization by actinomycetes has been associated with the use of (IUDs). Actinomyces may be seen in smears from secretions around the IUD, but has rarely been isolated in culture. Therefore there is no value in culturing these specimens

PURPOSE/RATIONALE:
To determine the presence or absence of *Actinomyces* spp. in intra-uterine devices (IUD) specimens.

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

SAMPLE INFORMATION:

Type	• IUD in a dry, sterile container
Stability	If the sample is received in the laboratory and processed greater than 24 hours from collection: • Add specimen quality comment: "Delayed transport may adversely affect pathogen recovery"
Storage Requirements	Room temperature
Criteria for rejection	1. Unlabeled/mislabeled swabs 2. Specimen container label does not match patient identification on requisition

REAGENTS and/or MEDIA:
- Brucella agar (BRU) and Thioglycollate broth (THIO)

SUPPLIES:

- Sterile red top vacutainer tube
- Sterile pipette
- Disposable inoculation needles
- Microscope slides
- Wooden sticks

EQUIPMENT

- Biosafety cabinet
- Vortex
- Centrifuge
- 35° ambient air and 35° CO_2 incubators
- 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 hand 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:

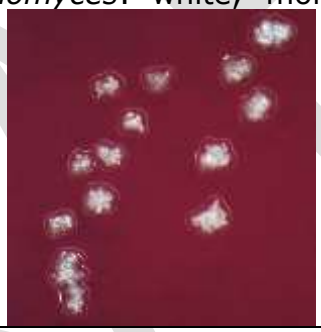
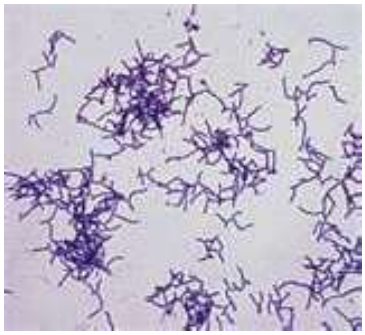
- Refer to Test Manual for reagent quality control procedures

PROCEDURE INSTRUCTIONS:

Step	Action
Processing specimens for IUD culture	
1	In the biosafety cabinet, add Thioglycollate broth to the specimen container containing the IUD and vortex for 30 seconds.
2	Using a sterile pipette, transfer the THIO broth to the red top vacutainer tube and centrifuge at 3500 rpm for 10 minutes.
3	After centrifugation is complete, remove the supernatant and: • Place 1 to 2 drops of sediment and BRU. Add 2 to 5 drops into THIO broth • Streak for isolated growth using a disposable inoculation needle

4	Incubate all media: • Place specimen and supernatant tube in the O_2 incubator • Label THIO with day 2 date, day 5 date and day 10 date and place in the THIO rack in the O_2 incubator • Place BRU in anaerobic jar with anaerobic pouch and indicator as soon as possible after inoculation. Label jar with day 2 date and place in the O_2 incubator **NOTE:** Anaerobes should not be exposed to air for 42 to 48 hours after inoculation

INTERPRETATION OF RESULTS:

Step	Action
1	• Observe BRU after 48 hours for growth of *Actinomyces* spp. • If no growth suggestive of *Actinomyces* spp. is observed, re-incubate BRU for an additional 72 hours • If no growth suggestive of *Actinomyces* spp. is observed at 5 days, re-incubate for an additional 5 days • After 5 days, examine plate for growth suggestive of *Actinomyces* spp. • Colonies typical of *Actinomyces*: white, "molar tooth," pitting the agar 
2	• Observe THIO after 48 hours for growth • If no growth in THIO is observed, re-incubate THIO for an additional 72 hours • If no growth is present in THIO at 5 days, re-incubate for an additional 5 days • If growth present, perform gram stain. if organisms resembling *Actinomyces* spp. are seen, culture broth to BRU agar and incubate anaerobically for 48 hours • Gram stain of *Actinomyces* spp. is branching, gram-positive bacilli 

3	From growth of colonies suggestive of *Actinomyces* app. on BRU, perform Vitek 2 ANC card to determine identification of organism.
4	From growth of subculture media from THIO, perform Vitek 2 ANC card to determine identification of organism.

REPORTING INSTRUCTIONS:

IF	REPORT
No *Actinomyces* spp. isolated	• Report: "**No Actinomyces isolated"**
Actinomyces spp. isolated	• Add organism**: "Actinomyces spp."** • List quantitation as **"Present"** • Report susceptibility results as per ASTM • Freeze isolate and log into stored isolates binder

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

APPROVAL:

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Date

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REVISION HISTORY:

REVISION	DATE	Description of Change	REQUESTED BY
1.0	20 Oct 17	Initial Release	L. Steven
2.0	30 Nov 18	Updated to include new Vitek 2 instrument	L. Steven
3.0	11 Jan 21	Procedure reviewed and added to NTHSSA policy template	L. Steven