PROGRAM Standard Operating Procedure – Laboratory Services	
Title: MIC32500 – Eye Culture-Superficial	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:
Inflammatory eye conditions can be separated into several clinical syndromes. A variety of microorganisms can play major roles, both in acute and in chronic conditions.

PURPOSE/RATIONALE:
This standard operating procedure describes how to determine the significance of growth in superficial eye specimens.

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

SAMPLE INFORMATION:

Type	Swab • Amie's with or without charcoal
Source	Conjunctiva: inflammation of the conjunctiva (the mucous membrane covering the sclera) ➢ Swab of conjunctiva surface / pus
Stability	If the sample is received in the laboratory and processed greater than 48 hours from collection: • Add specimen quality comment: "Delayed transport may adversely affect pathogen recovery"
Storage Requirements	Room temperature

Criteria for rejection	1. Unlabeled/mislabelled swabs 2. Specimen container label does not match patient identification on requisition

NOTE:

- If gonorrhoeae culture is ordered on eye specimen, superficial eye culture along with gonorrhoeae culture will be performed. Refer to MIC33500-Gonorrhoeae Culture
- Refer to MIC34100-Body Fluid Culture for intraocular fluid
- Refer tissue or biopsy specimens for culture to *Dyna*LIFE

REAGENTS and/or MEDIA:

- Blood agar (BA) and Chocolate agar (CHO)
- Identification reagents: catalase, oxidase, Staph latex test, Strep latex test, etc.

SUPPLIES:

- Disposable inoculation needles
- Microscope slides
- Wooden sticks

EQUIPMENT:

- Biosafety cabinet
- 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 potentially 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 superficial eye culture	
1	In the biosafety cabinet: • Inoculate BA and CHO with the swab • Ensure all surfaces of swab make contact with the agar • Streak for isolated growth using a disposable inoculation needle • Prepare smear by rolling the swab gently across the slide to avoid destruction of cellular elements and disruption of bacterial arrangements
2	Incubate the media: • Place BA and CHO in the CO_2 incubator
3	Allow smear to dry and perform gram stain. Gram stain must be read before culture plates. Refer to MIC20115-Gram stain procedure.

Probable Pathogens	
• *Haemophilus influenzae* • *Moraxella* spp. • *Pseudomonas aeruginosa* • *Neisseria gonorrhoeae*	• *Neisseria meningitidis* • *Staphylococcus aureus* • *Streptococcus pneumoniae* • *Streptococcus pyogenes*
Potential Pathogens	
• Anaerobes • Enteric Gram-negative bacilli • *Enterococcus* spp.	• *Haemophilus parainfluenzae* • Non-pathogenic *Neisseria* spp.
Commensal Skin Flora	
• Coagulase-negative *Staphylococcus* • *Corynebacterium* spp.	• *Micrococcus* spp. • viridans *Streptococcus* grp.

INTERPRETATION OF RESULTS:

Step	Action
1	Ensure growth on culture media correlates with gram stain results. If discordant results are found between the gram stain and growth: • Re-examine smear and culture plates • Check for anaerobic growth • Re-incubate media to resolve • Consider re-smearing or re-planting specimen
2	• Observe BA and CHO at 24 hours and 48 hours
3	• <u>If organism is a probable pathogen</u>: ➢ Perform and report full identification ➢ Perform and report susceptibility testing as per ASTM
4	• <u>If organism is a potential pathogen</u>: ➢ Perform and report full identification if there are ≤3 different potential pathogens ➢ Perform and report susceptibility testing as per ASTM on potential pathogens if ANY of the following is true:

	○ 3 to 4+WBC in the gram stain ○ Clinical diagnosis is infection ○ Patient is immunocompromised ○ Multiple cultures are positive for the same organism ➢ If >3 potential pathogens are present, list and do not perform or report susceptibility testing **NOTE:** Mixed enteric Gram-negative rods should be reported as mixture of coliform organisms, not reported individually
5	• If organism is commensal skin flora: ➢ Perform minimal identification and list **NOTE:** Mixed commensal skin flora should be reported as mixture of skin flora and not reported individually

REPORTING INSTRUCTIONS:

IF	REPORT
No growth after 1 day	**PRELIM:** • Report: **"No Growth After 1 Day"** • Report: **"Further report to follow"**
No growth after 2 days	**FINAL:** • Report: "**No Growth After 2 Days"**
Growth of probable pathogen	• Report organism full identification • List quantitation • Report susceptibility results as per ASTM
Growth of potential pathogen where full identification is required	• Report organism full identification • List quantitation • If indicated by procedure, perform and report susceptibility testing as per ASTM
Growth of potential pathogens where minimal identification and listing is required	• Report the minimal identification (i.e., Gram Negative Bacilli - Lactose Fermenter) • List quantitation
Growth of commensal skin flora where minimal identification and listing is required	• Report the minimal identification (i.e., Coagulase negative Staphylococci) • List quantitation
Mix of commensal skin flora	• Report: "**Mixture of skin flora"** • List quantitation
Mix of enteric Gram-negative bacilli	• Report: **"Mixture of coliform organisms"** • List quantitation

NOTE:

- Refer to Reportable Diseases – Public Health Act as of September 2009 for reporting to OCPHO (HPU1)
- Refer to LQM70620-Laboratory Critical Results List-Microbiology for results that need to be phoned to ordering location
- Refer to MIC36100-Nosocomial Infection Notification Job Aid to determine if organism needs to be copied to Infection Prevention and Control
- Refer to MIC36200-Referral of Category A Specimens to APL for sending category A isolates to APL
- Refer to MIC36300-Referral of Category B Specimens to APL for sending isolates to APL
- Refer to MIC36400-Referral of Category B Specimens to DL for sending isolates to *Dyna*LIFE.

LIMITATIONS:

1. False positive cultures can result from contamination of the specimen or plates with skin flora.
2. False negative results can occur if antimicrobial agents are given prior to collection of the specimen.
3. Even with the best techniques, culture often fails to yield the infecting organism.

CROSS-REFERENCES:

- LQM70620-Laboratory Critical Results List-Microbiology
- MIC20115-Gram stain procedure
- MIC33500-Gonorrhoeae Culture
- MIC34100-Body Fluid Culture
- MIC36100-Nosocomial Infection Notification Job Aid
- MIC36200-Referral of Category A Specimens to APL
- MIC36300-Referral of Category B Specimens to APL
- MIC36400-Referral of Category B Specimens to DL

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	28 May 18	Initial Release	L. Steven
2.0	26 Feb 21	Procedure reviewed and added to NTHSSA policy template	L. Steven
3.0	27 Feb 23	Procedure reviewed	L. Steven