


STANTON TERRITORIAL HEALTH AUTHORITY

Yellowknife, Northwest Territories

TITLE: Disc Diffusion-Optochin	Revision Date: 20-April-2018	Issue Date: 20-April-2016
Document Number: MIC50630	Status: Approved	
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Approved by: S. Asmussen, Manager of Diagnostic Services	Signed by: 	

PURPOSE:

For the differentiation of *Streptococcus pneumoniae* from other alpha-hemolytic streptococci.

SAMPLE INFORMATION:

Type	Pure, fresh culture of alpha gram positive organism
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REAGENTS and/or MEDIA:

Media	Information
Blood Agar (BAP)	<ul style="list-style-type: none">▪ Source: Oxoid▪ Store at 2 – 8°C away from direct light
Optochin disc (5µg) 6mm in diameter	<ul style="list-style-type: none">▪ Oxoid Canada▪ Storage Conditions -20 °C to 8 °C<ul style="list-style-type: none">• Store unopened cartridges in freezer• Opened cartridges can be kept in refrigerator at 2-8 °C (Oxoid Canada, 2008)

SUPPLIES:

- Sterile loop
- Tweezers
- Ruler

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SPECIAL SAFETY PRECAUTIONS:

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

- 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 where there is a known or potential risk of exposure to 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.

Optochin discs have the possibility of causing allergic sensitization; apply the use of proper personal protective equipment as outlined in Optochin Disc MSDS sheet.

QUALITY CONTROL:

Quality Control is set up each day the test is performed using the following control organisms. A QC order is generated in the TQC system.

Positive: *Streptococcus pneumoniae* ATCC # 49619
Negative: *Streptococcus viridans* ATCC # 13419

- A TQC order is automatically generated to record the QC results

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PROCEDURE INSTRUCTIONS:

Step	Action
Testing For Optichin Sensitivity	
1	In the plate log – Order ^TXP
2	Streak a pure culture of the organism across a BA plate
3	Apply an Optochin Disc to an area of heavy inoculum
4	Incubate in CO2 incubator for 16-18 hrs
5	Examine plate for a zone of inhibition.

INTERPRETATION:

IF	THEN
Zone size 6-8mm	▪ <u>NOT</u> <i>Streptococcus pneumonia</i>
Zone size 9-13mm	▪ <u>CANNOT RULE-OUT</u> <i>Streptococcus pneumoniae</i> as it could be an Optochin resistant strain. ▪Repeat test. ▪Perform Bile Solubility testing – See Procedure MIC52900
Zone size greater than or equal to 14mm	▪Report <i>Streptococcus pneumoniae</i>

REFERENCES:

- American Society For Microbiology. (2004). Clinical Microbiology Procedures Handbook. In H. D. Isenberg (Ed.).
- Murray, P., Baron, E. J., Joregensen, J., Landry, M. L., & Pfaller, M. (2007). *Manual of Clinical Microbiology*. Washington, DC, USA: ASM Press.
- Oxoid Canada. (2008). Diagnostic Discs Optochin Discs. *DD0001* .
- Oxoid Canada. (2008, October 07). Diagnostic Discs-Material Safety Data Sheet. *Safety Data Sheet Diagnostic Discs* .

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REVISION HISTORY:

REVISION	DATE	Description of Change	REQUESTED BY
1.0	15-NOV-2010	Initial Release	M-L Dufresne
1.1	31-Dec-2013	LIS Updates	A.Darrach
2.0	31-Mar-2016	Update of "Special Safety Precautions" to reflect risk assessment recommendations.	C. Russell