




STANTON TERRITORIAL HEALTH AUTHORITY

Yellowknife, Northwest Territories

TITLE: Oxidase	Revision Date: 20-April-2018	Issue Date: 20-April-2016
Document Number: MIC51400	Status: Approved	
Distribution: Microbiology Test Manual	Page: 1 of 4	
Approved by: S. Asmussen, Manager of Diagnostic Services	Signed by: 	

INTRODUCTION:

This test is useful in differentiating organisms, especially member of genera *Neisseria*, *Pseudomonas*, *Moraxella*, *Vibrio* and *Aeromonas* which are oxidase positive.

PRINCIPLE:

This test detects the presence of intracellular oxidase enzymes (cytochromes) which play a part in the electron transport system of respiration in some aerobic and facultative bacteria. The reagent is based upon the oxidation of tetramethyl-p-phenylenediamine by bacterial cytochromes in the presence of atmospheric oxygen to form a **purple** coloured compound (Wurster's **blue**).

SAMPLE INFORMATION:

Type	One well isolated colony
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REAGENTS and/or MEDIA:

Type	Pro-Lab Test Oxidase Reagent, Cat#PL.390
Storage Requirements	<ul style="list-style-type: none">• Store at controlled room temperature (15°C – 30°C) in the original container.• Do not freeze or overheat.• Protect from light.• Keep the screw cap tightly closed.
Stability	Product stored under the above conditions will be stable until expiry date shown on the label.

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SUPPLIES:

- Inoculating loop or sticks
- Filter paper strips or pads
- Incubator
- QC organisms

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.

QUALITY CONTROL:

Quality Control is set up each day the test is performed using the following control organisms:

Positive: *Pseudomonas aeruginosa* ATCC # 27853

Negative: *Klebsiella pneumoniae* ATCC # 13883

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

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PROCEDURE INSTRUCTIONS:

Step	Action
Direct Colony Method	
1	In the plate log – Order ^OX
2	Add one drop of reagent to a well-isolated colony on the surface of recommended agar medium.
3	Observe the colony for a colour change within 30 seconds. (If test isolate produces excessively mucoid or slimy colonies, allow up to 1 minute for colour development.)

Step	Action
Filter Paper Method	
1	In the plate log – Order ^OX
2	Add 1 to 2 drops of reagent to any convenient size of filter paper. Wait 1 to 2 minutes for proper reagent redistribution.
3	Using a wooden mixing stick or disposable inoculating loop (nichrome wire loops are not recommended), remove a medium size colony from the surface of the recommended agar medium and rub the inoculum onto the reagent-saturated area of the filter paper.
4	Observe the filter paper for colour change within 30 seconds.

INTERPRETATION OF RESULTS:

Positive test:

The production of a distinct blue or **purple** colour.

Negative test:

The absence of a distinct **blue** or **purple** colour.



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NOTES AND PRECAUTIONS:

1. Avoid contact with skin, eyes and clothing. Flammable.
2. The reagent should appear colourless, cloudy or very **light tan**. Do not use if the reagent is **purple**.
3. Do not take organisms off of media with dyes or indicators such as MacConkey since it will interfere with the colour reaction.

REFERENCES:

- Test Oxidase Reagent package insert, 2009
- Clinical Microbiology Procedures Handbook Henry D. Isenberg – Editor in Chief 2004

REVISION HISTORY:

REVISION	DATE	Description of Change	REQUESTED BY
1.0	25/01/11	Initial Release	J. Whitson
1.1	31Jul13	Addition of Computer Steps and Illustration	A. Darrach
1.2	11Mar14	Changed from Document control number MTE11400 to MIC51400	C. Russell
2.0	31Mar16	Update of "Special Safety Precautions" to reflect risk assessment recommendations.	C. Russell

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