

70439 Oxidase Test

Oxidase Test is an important differential procedure which should be performed on all gram-negative bacteria that are to be identified.

Composition:

(1 package contains 50 disks)

disks contains N,N-dimethyl-p-phenylenediamine oxalate and α -naphthol

Directions:

Take a inoculating loop or a toothpick. Then touch and spread a well isolated colony on a oxidase disk. The reaction is observed within 2 minutes at 25-30°C.

Principle and Interpretation:

Gordon and McLeod (1) introduced oxidase test for identifying Gonococci based upon the ability of certain bacteria to produce indophenol blue from the oxidation of dimethyl-p-phenylenediamine and α -naphthol. Gaby and Hadley (2) introduced amore sensitive method by using N,N-dimethyl-p-phenylenediamine oxalate where all Staphylococci were oxidase negative.

In presence of the enzyme cytochrome oxidase (gram-negative bacteria) the N,N-dimethyl-p-phenylenediamine oxalate and α -naphthol react to indophenol blue.

Oxidase test is mainly used to differentiate:

- 1) Oxidase positive *Neisseria* from other gram-negative diplococci.
- 2) Oxidas positive *Aeromonas hydrophila* from *Escherichia coli* (gram-negative)
- 3) Oxidase positive *Plesiomonas shigelloids* from *Shigella sonnei* (gram-negative)

Note: Cytochrome oxidase production may be inhibited by acid production and false negative reaction may be given by *Vibrio*, *Aeromonas*, and *Plesimonas* species when grow on a medium containing fermentable carbohydrate e.g. MacConkey Agar (Fluka 70143). Colonies taken from media containing nitrate may give unreliable results. The loss of activity of the oxidase reagent is caused by auto oxidation which may be avoided by adding 0.1% ascorbic acid (Fluka 95209).

Reaction within 2 minutes at 25-35°C.

Organisms (ATCC)	Reaction	Color
<i>Pseudomonas aeruginosa</i> (27853)	positive	deep purple blue
<i>Staphylococcus aureus</i> (25923)	negative	-
<i>Neisseria gonorrhoeae</i> (19424)	positive	deep purple blue
<i>Escherichia coli</i> (25922)	negative	-

References:

4. J.Gordon, J.W. McLeod, J. Path.Bact., 31, 185 (1928)
5. W.L. Gaby, C. Hadley, J. Bact., 74, 365 (1957)
6. K.J. Steel, J. Appl. Bact., 25, 445 (1962)