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Section: Technical Manual	Subject Title: Catalase Test	
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CATALASE TEST

Principle

Detects the presence of the enzyme catalase which hydrolyzes H₂O₂ to produce H₂O and O₂. This test is used to differentiate Staphylococci (catalase positive) from Streptococci (catalase negative).

Reagents

Hydrogen peroxide (H₂O₂), 3%

Store in a dark bottle and avoid any undue exposure to light.

Keep refrigerated at all times when not in use.

Other Materials

Clean glass microscope slides

Plastic culture loop or wooden applicator stick

Procedure

1. Pick a colony from an 18-24 hr culture and place it on a clean glass slide. Avoid carry over of blood agar which can cause false positives.
2. Put one drop of 3% H₂O₂ over the organism on the slide. Do not reverse the order of the procedure as false positive results may occur. Do not mix.
3. Observe for immediate bubbling (gas liberation) and record the result.
4. Discard the slide into a discard container.

Interpretation

Positive test: Immediate bubbling, easily observed (O₂ formed)

Negative test: No bubbling

Precautions

1. Carry over of blood agar must be avoided.
2. Growth for testing must be from an 18-24 hr culture.
3. 3% H₂O₂ is caustic - avoid exposure to skin. If H₂O₂ does get on the skin, immediately flood the area with 70% ethyl alcohol, not water.

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4. Aerosols may be released by the bubbling of the O₂.
5. H₂O₂ is unstable and breaks down easily on exposure to light. The solution must be kept refrigerated in the dark.

Quality Control

H₂O₂ is very unstable and should be tested daily or immediately prior to its use.

Positive: *S. aureus* (ATCC 25923)
 Negative: Gp. A. Strep. (ATCC 19615)

References

1. MacFaddin JF, Biochemical Tests for Identification of Medical Bacteria, 2nd ed., Williams and Wilkins, Baltimore MD., 1980, p51-58.
2. Murray PA, et al. Manual of Clinical Microbiology, 7th ed., 1999; pp 426-427.