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Section: Technical Manual	Subject Title: Streptococcal Grouping	
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STREPTOCOCCAL GROUPING

Principle

This test is used to determine the Lancefield group of an isolate. Latex particles labelled with specific group antisera will agglutinate in the presence of the corresponding antigen after extraction with nitrous acid.

Reagents

Pro-lab Streptococcal grouping Latex kit.

Other Materials

Droppers
Disposable slides
Wooden stirring sticks
13x100 mm test tubes

Procedure

- 1. Label one test tube for each isolate.
- 2. Add one drop of Extraction Reagent 1 to each tube.
- 3. Suspend 4 beta-haemolytic colonies in the Extraction Reagent 1.
- 4. Add 1 drop of Extraction Reagent 2 to each tube.
- 5. Shake the tube and incubate for 2 minutes at RT.
- 6. Add 7 drops of Extraction Reagent 3 to each tube. Mix the reaction by vortexing the tube for 10 15 seconds.
- 7. Dispense one drop of each latex suspension to be tested onto separate circles on the test card.
- 8. Using a pasteur pipette, add one drop of extract to the latex suspension.
- 9. Mix the latex and extract with the wooden stick using the complete area of the circle.
- 10. Gently rock the card for 2 minutes and look for agglutination.

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Interpretation

Positive: Strong visible agglutination within 2 minutes.

Negative: Milky appearance without visible agglutination.

Precautions

1. False positive reactions have been known to occur with organisms from unrelated genera eg. *E. coli, Klebsiella* sp., *Pseudomonas* sp.

Quality Control

Test reagents are checked weekly.

Each test should be tested with at least one extra grouping latex suspension as a negative control.

Reference

1. Pro-lab Streptococcal Grouping package insert.