

Procedures: Orion Staph Latex™

Prepared by	Date Adopted	Supersedes Procedure #

Review Date	Revision Date	Signature

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PRINCIPLE:

Traditionally, differentiation between coagulase positive and negative staphylococci has been performed with the tube coagulase test which detects extracellular staphylocoagulase⁽¹⁾ or the slide coagulase test that detects clumping factor (bound coagulase)⁽²⁾ present on the bacterial cell surface.

The demonstration of protein A in over 95% of the human strains of *S. aureus*⁽³⁾ has provided a basis for a slide procedure using latex particles coated with human plasma⁽⁴⁾. The high affinity of protein A to the Fc portion of immunoglobulin G (IgG), and of bound coagulase for fibrinogen, allows rapid agglutination of the coated latex particles in the presence of *S. aureus*.

SPECIMEN:

Use only colonies morphologically and biochemically resembling staphylococci (Gram positive cocci in clusters, and catalase positive). Fresh cultures grown overnight on media such as blood agar or nutrient agar give the best results. Collect the equivalent of four 1mm colonies from a primary or secondary culture plate. In case of insufficient growth, a subculture should be made. If sampling from a DNase agar make certain that the colonies used are not covered with hydrochloric acid.

EQUIPMENT AND MATERIALS:

Materials:

Provided in kit:

Orion Staph Latex Reagent

1 bottle containing 6 ml of a buffered suspension of modified latex particles covalently bound to purified human fibrinogen and human IgG.

Storage Requirements:

The Orion Staph Latex reagent should be stored at 2-8°C upon arrival. Do not freeze. The latex particles should be mixed carefully before use to get a homogeneous suspension and need not be brought to room temperature before use. Do not leave the latex reagent at room temperature for more than 1 hour. Discard the latex reagent if autoagglutination is present or if the expiration date is exceeded.

QUALITY CONTROL:

Daily quality control using a known strains of *S. aureus* such as ATCC 29213, and *S. epidermidis* ATCC 12228 are recommended as positive and negative controls, respectively. Inspect the latex suspension for granularity as it is placed on the test slide. Autoagglutination or granularity indicates contamination, freezing or deterioration of the reagent.

PROCEDURE - STEPWISE:

1. Gently mix the Orion Staph Latex reagent and dispense one drop directly onto a slide.
2. Pick approximately four suspect Staph colonies and emulsify directly into the latex reagent. Be careful not to damage the surface of the slide when mixing.
3. Rotate the slide until visible agglutination and a substantially clear background is observed, but not longer than 60 seconds. The agglutination patterns are clear cut and recognized under normal lighting conditions.

REPORTING RESULTS:

A visible clumping with substantial clearing of the Orion Staph Latex reagent indicates a POSITIVE reaction. In most cases the reaction is instantaneous. Rough or smooth filamentous reactions with clear background also indicate positive reaction. The test is considered NEGATIVE when no agglutination occurs or when there is little change in the milky appearance of the reagent. Filamentous agglutinates as well as fine agglutinates with a milky background indicate a negative reaction.

LIMITATIONS OF THE PROCEDURE:

Specimens collected from media with a high salt concentration such as mannitol-salt agar may have reduced protein A or clumping factor, and may cause false negative or relatively weak reactions.

Certain strains of *S. hyicus* and *S. intermedius* that produce coagulase⁽⁵⁾ may react with this test. However they are rarely isolated from human specimens. *S. saprophyticus* may give a weak positive result. Further biochemical testing may be required on these isolates for confirmation.

Certain yeast and other bacteria may also react nonspecifically with the reagent. A Gram stain on these suspect specimens is advised.

REFERENCES:

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