

Trypticase™ Soy Agar with 5% Sheep Blood

PRODUCT INFORMATION

I INTENDED USE

Trypticase Soy Agar with 5% Sheep Blood is extensively used for the recovery and cultivation of fastidious microbial species and for the determination of hemolytic reactions which are important differentiating characteristics for bacteria, especially *Streptococcus* species.

II SUMMARY AND EXPLANATION

The nutritional composition of **Trypticase Soy Agar** has made it a popular medium, both unsupplemented and as a base for media containing blood.

III PRINCIPLES OF THE PROCEDURES

The combination of casein and soy peptones in the **Trypticase Soy Agar** base render the medium highly nutritious by supplying organic nitrogen, particularly amino acids and larger-chained peptides. The sodium chloride maintains osmotic equilibrium.

Trypticase Soy Agar with 5% Sheep Blood (TSA II) provides excellent growth and beta hemolysis by *Streptococcus pyogenes* (Lancefield group A) and also provides excellent growth and appropriate hemolytic reactions with other fastidious organisms. It is suitable for performing the CAMP test for the presumptive identification of group B streptococci (*S. agalactiae*), and for use with low concentration (0.04 unit) bacitracin disc (**Taxo™A**) for presumptive identification of group A streptococci (*S. pyogenes*).

IV PROCEDURE

Instructions

The agar surface should be smooth and moist, but without excessive moisture.

Streak the specimen as soon as possible after it is received in the laboratory. The streak plate is used primarily to isolate pure cultures from specimens containing mixed flora.

Alternatively, if material is being cultured directly from a swab, roll the swab over a small area of the surface at the edge, then streak from this inoculated area.

Since many pathogens require carbon dioxide on primary isolation, plates may be incubated in an atmosphere containing approximately 3 to 10% CO₂.

Incubate plates in an aerobic atmosphere at 35 ± 2°C for 18 to 24 h.

Do not incubate anaerobically or in a CO₂ incubator. False-positive results may occur with group A streptococci when incubation is in an anaerobic or CO₂-enriched atmosphere.

V RESULTS

After incubation most plates will show an area of confluent growth. Because the streaking procedure is, in effect, a “dilution” technique diminishing numbers of microorganisms are deposited on the streaked areas. Consequently, one or more of these areas should exhibit isolated colonies of the organisms contained in the specimen. Further, growth of each organism may be semi-quantitatively scored on the basis of growth in each of the streaked areas.

1. Hemolytic streptococci may appear as translucent or opaque, grayish, small (1 mm), or large matt and mucoid (2 to 4 mm) colonies, encircled by a zone of hemolysis. Gram stains should be made and examined to check the macroscopic findings. (Other organisms which may cause hemolysis include *Listeria*, various corynebacteria, hemolytic staphylococci, *Escherichia coli* and *Pseudomonas*.)
In reporting, approximate quantitation of the number of colonies of hemolytic streptococci may be helpful to the clinician.
2. Pneumococci usually appear as very flat, smooth, translucent, grayish and sometimes mucoid colonies surrounded by a narrow zone of “green” (alpha) hemolysis.
3. Staphylococci appear as opaque, white to gold-yellow colonies with or without zones of beta hemolysis.
4. *Listeria*. Small zones of beta hemolysis are produced. They may be distinguished by their rod shape in stains, and by motility at room temperature.
5. Other organisms representing minimal flora and clinically significant isolates can also be expected to grow on this nonselective formulation.