BD™ Group A Selective Strep Agar with 5% Sheep Blood (ssA™)

INTENDED USE

BD Group A Selective Strep Agar with 5% Sheep Blood (**ssA**™) is a selective medium for use in the isolation and presumptive identification of group A streptococci from throat cultures and other clinical specimens.

PRINCIPLES AND EXPLANATION OF THE PROCEDURE

Microbiological method.

Infection with Lancefield group A streptococci (*Streptococcus pyogenes*) may produce serious sequelae such as rheumatic fever and acute glomerulonephritis. Therefore, early detection and identification are important. Because of the overgrowth of normal flora present in throat culture specimens plated on routine blood agar plates, selective ingredients have been added to sheep blood agar to enhance the detection of group A streptococci.

Evaluation of various antimicrobial agents in our laboratories resulted in a combination with improved selectivity over other selective media tested. This medium (ssA) allows presumptive identification of group A streptococci, based on bacitracin susceptibility and beta hemolysis, within 24 h after inoculation with the specimen when the medium is incubated in a carbon dioxide-enriched atmosphere.[1,2]

BD Group A Selective Strep Agar with 5% Sheep Blood incorporates a unique combination of selective ingredients in **Trypticase™ Soy Sheep Blood II Agar** (TSA II) to suppress normal throat flora for improved recovery of *S. pyogenes*. Defibrinated sheep blood supplies enrichment for the growth of such fastidious organisms and allows detection of the typical beta-hemolysis of S. pyogenes. Beta-hemolytic streptococci which show a zone of inhibition around a bacitracin (0.04 unit) disc may be presumptively identified as group A streptococci.

REAGENTS

BD Group A Selective Strep Agar with 5% Sheep Blood (ssA)

Formula* Per Liter Purified Water

Pancreatic Digest of Casein	14.5 g
Papaic Digest of Soybean Meal	5.0
Sodium Chloride	5.0
Agar	14.0
Growth Factors	1.5
Selective Agents	40.2 mg
Sheep Blood, defibrinated	5 %

pH 7.4 ± 0.2

*Adjusted and/or supplemented as required to meet performance criteria.

PRECAUTIONS

IVD . For professional use only.

Do not use plates if they show evidence of microbial contamination, discoloration, drying, cracking or other signs of deterioration.

Consult **GENERAL INSTRUCTIONS FOR USE** document for aseptic handling procedures, biohazards, and disposal of used product.

STORAGE AND SHELF LIFE

On receipt, store plates in the dark at 2 to 8° C, in their original sleeve wrapping until just prior to use. Avoid freezing and overheating. The plates may be inoculated up to the expiration date (see package label) and incubated for the recommended incubation times.

Plates from opened stacks of 10 plates can be used for one week when stored in a clean area at 2 to 8° C.

USER QUALITY CONTROL

Inoculate representative samples with the following strains (for details, see **GENERAL INSTRUCTIONS FOR USE** document). A **BD Taxo**™ A disc (0.04 U of bacitracin per disc) may be placed at the intersection of the first and second area of streaking on all plates inoculated with S. pyogenes.
Incubate plates at 35 ± 2°C in an aerobic atmosphere supplemented with carbon dioxide. Examine plates after 18 to 24 h for beta hemolysis and for amount of growth, inhibition, colony size and hemolytic reactions.

Strains	Growth Results
Streptococcus pyogenes ATCC™ 19615	Good to excellent growth; tiny to small colonies with ß-hemolysis; inhibition zone around a bacitracin disc
Streptococcus mitis DSM 12643	Inhibition partial to complete
Staphylococcus aureus ATCC 25923	Inhibition complete
Neisseria subflava ATCC 14799	Inhibition complete
Escherichia coli ATCC 25922	Inhibition partial to complete
Uninoculated	Red to dark red (blood color)

PROCEDURE

Materials Provided

BD Group A Selective Strep Agar with 5% Sheep Blood (ssA), provided in 90 mm **Stacker™** plates. Microbiologically controlled.

Materials Not Provided

Ancillary culture media, reagents and laboratory equipment as required.

Specimen Types and Collection

This medium is used for throat specimens or any other specimens suspected to contain *Streptococcus pyogenes* (=A streptococci).
Throat specimens suitable for culture may be obtained by swabbing the pharynx and tonsillar area of the throat with a polyester- or polyurethane-tipped swab, taking care to avoid touching the tongue or uvula. Sources other than the throat should be cultured according to recommended procedures. Appropriate transport media should be used for delayed transport. For detailed information, appropriate texts should be consulted.[3,4]

Test Procedure

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. A **BD Taxo** A disc (0.04 U of bacitracin per disc) may be placed at the intersection of the first and second area of streaking on all plates. A non-selective blood agar plate such as **BD Columbia Agar with 5% Sheep Blood** ot **BD Trypticase Soy Agar II with 5% Sheep Blood** should also be inoculated to detect other pathogens potentially present in the specimen.
Incubate inoculated plates at 35 ± 2°C in an atmosphere enriched with carbon dioxide. Examine plates after 18 to 24 h.

Results

After 18 to 24 h of incubation in an atmosphere enriched with carbon dioxide, group A streptococci (*S. pyogenes*) on **ssA** will appear as translucent or opaque, white to gray, small (1 to 2 mm) colonies surrounded by a zone of beta hemolysis. A decrease in size as compared to the nonselective control, Trypticase Soy Agar with 5% Sheep Blood, is typical. Pinpoint or very small colonies of alpha-, nonhemolytic or other beta-hemolytic streptococci may grow in small

numbers, but they should not interfere with the recovery of group A streptococci or interpretation of the results. *Neisseria* species, viridans streptococci, staphylococci, gram-negative rods and most beta-hemolytic streptococci other than groups A and B are inhibited on the **ssA** medium. Bacitracin susceptibility may be used to differentiate group A streptococci from group B. Fair to heavy growth of beta-hemolytic colonies demonstrating a zone of inhibition around the **Taxo** A disc may be presumptively reported as *S. pyogenes.* A PYR (pyroglutamic acid) test may also be performed. It is more specific and as sensitive as the bacitracin test for this purpose.[3] Gram stains should be made and examined.
A serological grouping test procedure may be performed if sufficient well-isolated beta-hemolytic colonies are present.

PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE PROCEDURE

BD Group A Selective Strep Agar with 5% Sheep Blood (**ssA**) is suitable for use with specimens contaminated with normal flora such as thrat swabs or other (e.g., would swabs or pus) suspected to contain A streptococci.
In a performance evaluation consisting of 460 throat cultures, there was a total of 117 positive for group A streptococci (*S. pyogenes*) on the **BD Group A Selective Strep Agar with 5% Sheep Blood** (**ssA**) compared to 100 on SXT Sheep Blood Agar and 84 of **BD Trypticase Soy Agar with 5% Sheep Blood** (**TSA II**). Of these positive cultures, 103 were identified based on beta hemolysis and bacitracin (0.04 unit) susceptibility within 24 h on **ssA** compared with 80 on SXT and only 32 on the nonselective TSA blood agar control.[2]
BD Group A Selective Strep Agar with 5% Sheep Blood (**ssA**) does not inhibit all strains of B-streptococci (*Streptococcus agalactiae*).
Since there is no such entity as a perfect medium, some strains of group A streptococci (*S. pyogenes*) may be encountered that will grow poorly on this medium; the nature of the specimens and the physiologic state of the organisms can influence recovery of the desired species, as well as modify the effects of the inhibitory characteristics of the medium. It is therefore useful to compare the growth on this medium with the growth obtained on a non-selective blood agar, such as **BD Columbia Agar with 5% Sheep Blood** or **BD Trypticase Soy Agar II with 5% Sheep Blood** to obtain additional information and to assure optimal recovery of any potential pathogens.
Some diagnostic tests may be performed with the primary isolation plate. However, a pure culture is recommended for biochemical tests and serological procedures. Consult appropriate texts for detailed information and recommended procedures.[3-5]

REFERENCES

1. Evans, G.L., and T.E. O'Neill. 1984. Development of an improved selective medium for the isolation of group A streptococci from throat cultures, Abstr. C-136, p. 259. Abstr. 84th Annu. Meet. Am. Soc. Microbiol. 1984.
2. Carlson, J.R., W.G. Merz, B.E. Hansen, S. Ruth, and D.G. Moore. 1985. Improved recovery of group A beta-hemolytic streptococci with a new selective medium. J. Clin. Microbiol. 21:307-309.
3. Baron, E.J., L.R. Peterson, and S.M. Finegold. 1994. Bailey & Scott's diagnostic microbiology, 9th ed. Mosby-Year Book, Inc., St. Louis.
4. Ruoff, K.L. 1995. *Streptococcus*, p. 299-307. In P.R. Murray, E.J. Baron, M.A. Pfaller, F.C. Tenover, and R.H. Yolken (ed.), Manual of clinical microbiology, 6th ed. American Society for Microbiology, Washington, D.C.
5. Koneman, E.W., S.D. Allen, W.M. Janda, P.C. Schreckenberger, and W.C. Winn, Jr. 1992. Color atlas and textbook of diagnostic microbiology, 4th ed. J.B. Lippincott Co., Philadelphia.

PACKAGING/AVAILABILITY

BD Group A Selective Strep Agar with 5% Sheep Blood (ssA)
Cat. No. 254050 Ready-to-use Plated Media, cpu 20

FURTHER INFORMATION

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