INTENDED USE

BD Neomycin Agar with 5% Sheep Blood is a selective medium used for the isolation of group A and B streptococci.

PRINCIPLES AND EXPLANATION OF THE PROCEDURE

Microbiological method.
Lancefield group A streptococci (S. pyogenes) and group B streptococci (S. agalactiae) are medically important gram-positive beta-hemolytic bacteria. Blanchette and Lawrence added neomycin to sheep blood agar to enhance the detection of Lancefield group A and group B streptococci by suppressing the growth of other hemolytic organisms. Most staphylococci, Enterobacteriaceae, Neisseria, Listeria and some Pseudomonas species are inhibited. Streptococci other than group A and B may also grow on the medium.

Neomycin Agar + 5% Sheep Blood contains Trypticase™ Soy Agar II (TSA II) as a base medium. The defibrinated sheep blood supplies enrichment for growth of fastidious organisms. The incorporation of 30 µg/ml of neomycin in this medium provides suppression of normal flora for improved recovery of the group A and group B streptococci.

REAGENTS

BD Neomycin Agar with 5% Sheep Blood

Formula* Per Liter Purified Water

<table>
<thead>
<tr>
<th>Component</th>
<th>Amount</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pancreatic Digest of Casein</td>
<td>14.5g</td>
</tr>
<tr>
<td>Papaic Digest of Soybean Meal</td>
<td>5.0</td>
</tr>
<tr>
<td>Sodium Chloride</td>
<td>5.0</td>
</tr>
<tr>
<td>Agar</td>
<td>14.0</td>
</tr>
<tr>
<td>Growth Factors</td>
<td>1.5</td>
</tr>
<tr>
<td>Neomycin</td>
<td>0.03</td>
</tr>
<tr>
<td>Sheep Blood, defibrinated</td>
<td>5%</td>
</tr>
</tbody>
</table>

pH 7.3 +/- 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). 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.

<table>
<thead>
<tr>
<th>Strains</th>
<th>Growth Results</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Streptococcus pyogenes</em> ATCC™ 19615 (Group A)</td>
<td>Growth good to excellent; small colonies with beta hemolysis</td>
</tr>
<tr>
<td><em>Streptococcus agalactiae</em> ATCC 12386 (Group B)</td>
<td>Growth good to excellent; medium-sized colonies with beta hemolysis</td>
</tr>
<tr>
<td><em>Escherichia coli</em> ATCC 25922</td>
<td>Inhibition (partial to) complete</td>
</tr>
<tr>
<td><em>Staphylococcus aureus</em> ATCC 25923</td>
<td>Inhibition partial to complete, may be beta-hemolytic</td>
</tr>
<tr>
<td>Uninoculated</td>
<td>Red (blood color)</td>
</tr>
</tbody>
</table>

### PROCEDURE

#### Materials Provided

**BD Neomycin Agar with 5% Sheep Blood** (90 mm Stacker™ plates). Microbiologically controlled.

#### Materials Not Provided

Ancillary culture media, reagents and laboratory equipment as required.

#### Specimen Types

This medium may be used for all specimen types from patients suspected to have an infection with group A (= *Streptococcus pyogenes*) and group B (= *Streptococcus agalactiae*) streptococci. Specimens include, but are not limited to, throat swabs, vaginal swabs, or swabs collected from newborns (see also PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE PROCEDURE).

#### Test Procedure

As soon as possible after the specimen is received in the laboratory, inoculate the specimen onto **BD Neomycin Agar with 5% Sheep Blood** plate by firmly rolling swab over a third of the agar surface. Streak the remainder of the plate with a sterilized inoculating loop to obtain isolated colonies. Without re-sterilizing the loop, stab the agar two or three times in the area of heaviest inoculation. This will result in subsurface growth which will display the most reliable hemolytic reactions.

Also inoculate a **BD Columbia Agar with 5% Sheep Blood** plate or **BD Trypticase Soy Agar II with 5% Sheep Blood** plate to assure the recovery of other clinically significant microorganisms that may be inhibited on the selective medium.

Incubate inoculated plates at 35 ± 2°C in an aerobic atmosphere supplemented with carbon dioxide. Examine plates after 18 to 24 h and 48 h, if necessary.

#### Results

After 18 to 48 hours of incubation, group A and group B streptococci will appear as translucent or opaque, white to gray, small (1-2mm) colonies surrounded by a zone of beta hemolysis. A decrease in size as compared to the nonselective control is typical. In the areas where the medium has been stab-inoculated beta hemolysis will be clearer than around the colonies, if beta hemolytic streptococci are present.

*Neisseria* species, gram-negative rods and most beta-hemolytic staphylococci are inhibited. While both group A and group B streptococci will grow on this medium, susceptibility to bacitracin (0.04 unit), utilizing **BD Taxo™ A** discs, may be used to differentiate them. For details, refer to the respective Instructions for Use. A more reliable method for the identification of the Lancefield group is the use of a grouping kit for streptococci, or a biochemical identification system.

### PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE PROCEDURE

**BD Neomycin Agar with 5% Sheep Blood** is used for the selective isolation of group A and group B streptococci. Although the medium is selective and Gram negative rods are usually
inhibited, other organisms and other streptococci (e.g., *Streptococcus mitis*) may grow on this medium and may or may not produce beta hemolytic colonies. Therefore, biochemical or immunological tests are necessary for final identification of the isolates.

**BD Neomycin Agar with 5% Sheep Blood** is less selective than **BD Group A Selective Strep Agar (ssA™)** for the inhibition of normal throat flora. It is recommended that ssA medium is used when only group A streptococci are suspected.

**REFERENCES**

**Packaging/Availability**
**BD Neomycin Agar with 5% Sheep Blood**
Cat. No. 254444 Ready-to-use Plated Media, cpu 20

**FURTHER INFORMATION**
For further information please contact your local BD representative.

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