

# Instructions for Use

# SELECTIVE STREP AGAR

Cat. no. A70	Selective Strep Agar, 15x100mm Plate, 17ml	10 plates/bag

#### **INTENDED USE**

Hardy Diagnostics Selective Strep Agar is a selective medium recommended for use in the primary isolation of all *Streptococcus* species, including streptococcal groups A (*S. pyogenes*), B (*S. agalactiae*), C, D, F, G, and *S. pneumoniae*, especially from respiratory specimens.

# **SUMMARY**

Selective Strep Agar is designed to inhibit gram-negative bacilli and staphylococci, thereby allowing for the isolation, subculture, and identification of pathogenic streptococci, including beta-hemolytic streptococci and *S. pneumoniae*.

Tryptic Soy Agar is the basal medium for Selective Strep Agar. Organic nitrogen, particularly amino acids and long-chained peptides are supplied by the combination of casein and soy peptones. This combination renders the medium highly nutritious. Osmotic equilibrium is maintained by sodium chloride. Sheep blood (5%) has been added to facilitate growth and to detect hemolytic activity. Selective agents are added to suppress much of the oral flora, including coliforms, staphylococci, *Micrococcus*, *Haemophilus* and *Neisseria* species. All species of *Streptococcus* will grow on this medium. For the selection of only group A streptococci (*S. pyogenes*) see Group A Beta Strep Agar, Cat. no. A72.

# **FORMULA**

Ingredients per liter of deionized water:\*

Pancreatic Digest of Casein	15.0gm
Peptic Digest of Soybean Meal	5.0gm
Sodium Chloride	5.0gm
Nucleic Acid	3.0gm
Selective Agents	15.3mg
Sheep Blood	50.0ml
Agar	15.0gm

Final pH 7.3 +/- 0.2 at 25°C.

#### STORAGE AND SHELF LIFE

<sup>\*</sup> Adjusted and/or supplemented as required to meet performance criteria.

Storage: Upon receipt store at 2-8°C. away from direct light. Media should not be used if there are any signs of deterioration (shrinking, cracking, or discoloration), hemolysis, contamination, or if the expiration date has passed. Product is light and temperature sensitive; protect from light, excessive heat, moisture, and freezing.

The expiration date on the product label applies to the product in its intact packaging when stored as directed. The product may be used and tested up to the expiration date on the product label and incubated for the recommended incubation times as stated below.

Refer to the document "Storage" for more information.

# **PRECAUTIONS**

This product may contain components of animal origin. Certified knowledge of the origin and/or sanitary state of the animals does not guarantee the absence of transmissible pathogenic agents. Therefore, it is recommended that these products be treated as potentially infectious, and handle observing the usual Universal Precautions for blood. Do not ingest, inhale, or allow to come into contact with skin.

This product is for *in vitro* diagnostic use only. It is to be used only by adequately trained and qualified laboratory personnel. Observe approved biohazard precautions and aseptic techniques. All laboratory specimens should be considered infectious and handled according to "standard precautions." Refer to the document "Guidelines for Isolation Precautions" from the Centers for Disease Control and Prevention.

For additional information regarding specific precautions for the prevention of the transmission of all infectious agents from laboratory instruments and materials, and for recommendations for the management of exposure to infectious disease, refer to CLSI document M29: *Protection of Laboratory Workers from Occupationally Acquired Infections*.

Sterilize all biohazard waste before disposal.

Refer to the document "Precautions When Using Media" for more information.

#### **PROCEDURE**

Specimen Collection: Infectious material should be submitted directly to the laboratory without delay and protected from excessive heat and cold. If there is to be a delay in processing, the specimen should be inoculated onto an appropriate transport media and refrigerated until inoculation. Consult listed references for information on specimen collection. (1-5)

Prepared media should be inoculated, incubated, and results recorded according to accepted procedures described in the listed reference texts. (1-5)

Method of Use: Medium should be brought to room temperature prior to inoculation. Inoculate according to standard microbiological procedures. Streak inoculum so as to obtain isolated colonies. Stab the medium several times with the inoculating loop in the area of heavy inoculation in order to create anaerobic conditions to stimulate maximum expression of beta-hemolysis. Incubate at 35°C. for 18-24 hours.

It is recommended to incubate plates either anaerobically or with increased  $CO_2$  (5-10%) in order to enhance the development of hemolytic zones of the pathogenic streptococci.<sup>(7)</sup>

Square Pill Pocket Plate: After inoculation, place one CO<sub>2</sub> tablet in the pill pocket and place plate in a sealed zip bag. Do not invert the plate. Proceed with incubation parameters as outlined above.

#### **LIMITATIONS**

It is recommended that biochemical, immunological, molecular, or mass spectrometry testing be performed on colonies from pure culture for complete identification of bacteria and/or fungi.

Selective Strep Agar is designed to grow all species of streptococci, which would include some normal respiratory

flora, such as Streptococcus viridans.

*Arcanobacterium haemolyticum* may be recovered on Selective Strep Agar, however the colonies will likely be obscured by other flora, thus Arcanobacterium Selective Agar is recommended for *A. haemolyticum* recovery.

Unless a provision is made to reduce oxygen tension, approximately 2% of group A streptococci may be missed if incubated aerobically. It is recommended that several stabs be made into the medium upon inoculation. (2) Incubation in increased  $CO_2$  or anaerobically is recommended.

Either anaerobic incubation or aerobic with stabs (see Method of Use) is the preferable atmosphere. (6)

Selective Strep Agar may be inhibitory to certain rare strains of streptococci.

Refer to the document "Limitations of Procedures and Warranty" for more information.

# MATERIALS REQUIRED BUT NOT PROVIDED

Standard microbiological supplies and equipment such as loops, other culture media, swabs, applicator sticks, incinerators, and incubators, etc., as well as serological and biochemical reagents, are not provided.

# **QUALITY CONTROL**

Hardy Diagnostics tests each lot of commercially manufactured media using appropriate quality control microorganisms and quality specifications as outlined on the Certificate of Analysis (CofA) and the CLSI document M22-A3 *Quality Assurance for Commercially Prepared Microbiological Culture Media*. The following microorganisms are routinely used for testing at Hardy Diagnostics:

Test Organisms	Inoculation Method*	Incubation			Results
Test Organisms		Time	Temperature	Atmosphere	Results
Streptococcus pyogenes ATCC® 19615	A	24hr	35°C	CO <sub>2</sub> **	Growth; beta-hemolysis
Streptococcus pneumoniae ATCC <sup>®</sup> 6305	A	24hr	35°C	CO <sub>2</sub> **	Growth; alpha-hemolysis
Escherichia coli ATCC® 25922	В	24hr	35°C	CO <sub>2</sub> **	Inhibited
Staphylococcus epidermidis ATCC <sup>®</sup> 12228	В	24hr	35°C	CO <sub>2</sub> **	Inhibited

<sup>\*</sup> Refer to the document "Inoculation Procedures for Media QC" for more information.

#### **USER QUALITY CONTROL**

End users of commercially prepared culture media should perform QC testing in accordance with applicable government regulatory agencies, and in compliance with accreditation requirements. Hardy Diagnostics recommends end users check for signs of contamination and deterioration and, if dictated by laboratory quality control procedures or regulation, perform quality control testing to demonstrate growth or a positive reaction and to demonstrate inhibition or a negative reaction, if applicable. Hardy Diagnostics quality control testing is documented on the certificate of analysis (CofA) available from Hardy Diagnostics Certificate of Analysis website. Also refer to the document "Finished Product Quality Control Procedures," and the CLSI document M22-A3 Quality Assurance for Commercially Prepared Microbiological Culture Media for more information on the appropriate QC procedures. See the references below.

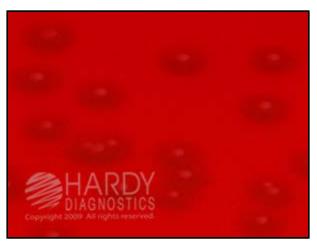
#### PHYSICAL APPEARANCE

<sup>\*\*</sup> Atmosphere of incubation is enriched with 5-10% CO<sub>2</sub>.

Selective Strep Agar should appear opaque, and cherry red in color.



Streptococcus pyogenes (ATCC $^{\circledR}$  19615) colonies growing on Selective Strep Agar (Cat. no. A70). Incubated in CO $_2$  for 24 hours at 35 $^{\degree}$ C.



Streptococcus pneumoniae (ATCC $^{\circledR}$  6305) colonies growing on Selective Strep Agar (Cat. no. A70). Incubated in CO $_2$  for 24 hours at 35°C.



Escherichia coli (ATCC® 25922) growth inhibited on Selective Strep Agar (Cat. no. A70). Incubated in CO<sub>2</sub> for 24 hours at 35°C.

# **REFERENCES**

- 1. Anderson, N.L., et al. *Cumitech 3B; Quality Systems in the Clinical Microbiology Laboratory*, Coordinating ed., A.S. Weissfeld. American Society for Microbiology, Washington, D.C.
- 2. Jorgensen., et al. Manual of Clinical Microbiology, American Society for Microbiology, Washington, D.C.
- 3. Tille, P., et al. Bailey and Scott's Diagnostic Microbiology, C.V. Mosby Company, St. Louis, MO.
- 4. Isenberg, H.D. *Clinical Microbiology Procedures Handbook*, Vol. I, II & III. American Society for Microbiology, Washington, D.C.
- 5. Koneman, E.W., et al. *Color Atlas and Textbook of Diagnostic Microbiology*, J.B. Lippincott Company, Philadelphia, PA.
- 6. The United States Pharmacopeia, XXII, 1990.

7. Pacifico, L., et al. 1995. Journal of Clinical Micro., 33; 9:2480-2482. American Society for Microbiology.

ATCC is a registered trademark of the American Type Culture Collection.

IFU-10749[B]



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