



## BETA-SSA AGAR (7336)

### Intended Use

**Beta-SSA Agar** is used with blood for the selective isolation of group A streptococci.

### Product Summary and Explanation

Group A streptococcal infections are the most common cause of bacterial pharyngitis in children 5 to 10 years old.<sup>1</sup> Beta-SSA Agar is a highly selective agar developed for the isolation of group A beta-hemolytic streptococci. The selective agents in the medium inhibit gram-negative bacteria and most Gram-positive bacteria, although some strains of group B beta-hemolytic streptococci may grow. Beta-SSA Agar is supplemented with 5% sheep blood to detect hemolytic patterns of streptococci.

### Principles of the Procedure

The nitrogen, vitamin, and carbon sources are provided by Enzymatic Digest of Casein, Enzymatic Digest of Soybean Meal and Enriched Peptone. Sodium Chloride maintains the osmotic balance of the medium. Selective Agents inhibit Gram-negative bacteria and most Gram-positive bacteria. Agar is the solidifying agent.

In general, blood agar bases are relatively free of reducing sugars, which have been reported to adversely influence the hemolytic reactions of  $\beta$ -hemolytic streptococci.<sup>2</sup> Supplementation with blood (5 - 10%) provides additional growth factors for fastidious microorganisms and aids in determining hemolytic reactions. Hemolytic patterns may vary with the source of animal blood and type of basal medium used.<sup>1</sup>

### Formula / Liter

Enzymatic Digest of Casein .....	15 g
Enzymatic Digest of Soybean Meal .....	2.5 g
Sodium Chloride .....	5 g
Enriched Peptone .....	2.5 g
Selective Agents .....	0.3002 g
Agar .....	15 g

Final pH: 7.3  $\pm$  0.2 at 25°C

Formula may be adjusted and/or supplemented as required to meet performance specifications.

### Precaution

1. For Laboratory Use.
2. IRRITANT. Irritating to eyes, respiratory system, and skin.

### Directions

1. Suspend 40 g of the medium in one liter of purified water.
2. Heat with frequent agitation and boil for one minute to completely dissolve the medium.
3. Autoclave at 121°C for 15 minutes.
4. Prepare 5% blood agar by aseptically adding the appropriate volume of sterile defibrinated blood to melted sterile agar medium, cooled to 45 - 50°C.

### Quality Control Specifications

**Dehydrated Appearance:** Powder is homogeneous, free flowing, and beige to light gray.

**Prepared Appearance:** Prepared medium with 5% sheep blood is red and opaque.

**Expected Cultural Response:** Cultural response on Beta-SSA Agar supplemented with 5% sheep blood at  $35 \pm 2^\circ\text{C}$  after 18 - 24 hours and 48 hours of incubation.

Microorganism	Approx. Inoculum (CFU)	Expected Results
<i>Escherichia coli</i> ATCC® 25922	$\geq 1000$	Inhibited
<i>Staphylococcus aureus</i> ATCC® 25923	$\geq 1000$	Inhibited
<i>Streptococcus pneumoniae</i> ATCC® 6305	$\geq 1000$	Inhibited
<i>Streptococcus pyogenes</i> ATCC® 19615	10 - 300	Hemolysis after 18 – 24 hours
<i>Streptococcus agalactiae</i> ATCC® 13813	10 - 300	Growth, no hemolysis @ 48 hours
<i>Streptococcus pyogenes</i> ATCC® 19615	Lawn (Bacitracin Differentiation Disk Test)	Any zone of inhibition after 18 – 24 hours

The organisms listed are the minimum that should be used for quality control testing.

### **Test Procedure**

1. Process each specimen as appropriate, and inoculate directly onto surface of the medium. Streak for isolation with inoculating loop, stab agar several times to deposit beta-hemolytic streptococci beneath the agar surface. Subsurface growth will display the most reliable hemolytic reactions owing to activity of both oxygen-stable and oxygen-labile streptolysins.<sup>1</sup>
2. Incubate plates aerobically, anaerobically, or under conditions of increased  $\text{CO}_2$  (5 - 10%) in accordance with established laboratory procedures.

### **Results**<sup>3</sup>

Examine medium for growth and hemolytic reactions after 18 - 24 and 48 hours incubation. Beta hemolysis ( $\beta$ ) is the lysis of red blood cells, producing a clear zone surrounding the colony.

### **Storage**

Store sealed bottle containing the dehydrated medium at 2 - 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light.

### **Expiration**

Refer to expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if appearance has changed from the original color. Expiry applies to medium in intact container.

### **Limitations of the Procedure**

1. Atmosphere of incubation is known to influence hemolytic reactions of beta-hemolytic streptococci.<sup>1</sup> For optimal performance, incubate blood agar base media under increased  $\text{CO}_2$  (5 - 10%).

### **Packaging**

<b>Beta-SSA Agar</b>	<b>Code No.</b>	<b>7336A</b>	<b>500 g</b>
		<b>7336B</b>	<b>2 kg</b>
		<b>7336C</b>	<b>10 kg</b>

### **References**

1. Murray, P. R., E. J. Baron, M. A. Pfaller, F. C. Tenover, and R. H. Tenover (eds.). Manual of clinical microbiology, 6<sup>th</sup> ed. American Society of Microbiology, Washington, D.C.
2. Casman, E. P. 1947. A noninfusion blood agar base for neisseriae, pneumococci and streptococci. Am. J. Clin. Pathol. 17:281-289.
3. Isenberg, H. D. (ed.). 1992. Interpretation of aerobic bacterial growth on primary culture media, Clinical microbiology procedures handbook, vol. 1 p. 1.61-1.67. American Society for Microbiology, Washington, D.C.

### **Technical Information**

Contact Acumedia Manufacturers, Inc. for Technical Service or questions involving dehydrated culture media preparation or performance at (517)372-9200 or fax us at (517)372-2006.