

## Product Insert

### BOTULISM SELECTIVE MEDIUM (BSM)

#### Products

AS-223 Botulism Selective Medium (BSM) 4 plates / pkg

The following product contains BSM as one of multiple components

AS-224 BSM / LVA 2 plates each / pkg

#### Intended Use

Botulism Selective Medium (BSM) is a selective and differential medium for the presumptive identification of *Clostridium botulinum* from food or clinical samples.

#### Summary

BSM agar is a selective and differential media used for the presumptive identification of *Clostridium botulinum* from food or fecal samples. BSM contains cycloserine, sulfamethoxazole, and trimethoprim as selective inhibitory agents. A suspension of egg yolk is added for the detection of lecithinase, lipase, and proteolytic activity. The degradation of lecithin in the egg yolk results in an opaque precipitate around the colonies. Lipase destroys the fats within the egg yolk, which results in an iridescent sheen on the colony surface. Proteolytic activity is indicated by a clearing of the medium around the colonies. This medium is dispensed under oxygen-free conditions to prevent the formation of oxidized products prior to use.

#### Formulation\*

|                         |       |    |
|-------------------------|-------|----|
| Beef Heart Infusion     | 10.00 | g  |
| Tryptose                | 10.00 | g  |
| Sodium Chloride         | 5.00  | g  |
| Cycloserine             | 0.25  | g  |
| Sulfamethoxazole        | 0.076 | g  |
| Trimethoprim            | 0.06  | g  |
| Thymidine Phosphorylase | 0.10  | mL |
| Egg Yolk Suspension     | 40.00 | mL |
| Agar                    | 20.00 | g  |
| DI Water                | 1.00  | L  |

Final pH: 7.4 ± 0.3 at 25 °C

Final weight: 17.0 g ± 1.7 g

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

#### Precautions

For *IN VITRO DIAGNOSTIC USE* only. Utilize approved biohazard precautions and aseptic technique when using this product. This product is for use only by properly-trained and qualified personnel. Sterilize all biohazard waste prior to disposal.

#### Storage and Shelf Life

**Storage:** Upon receipt, store at room temperature in original package until used. Avoid overheating or freezing. Do not use media if there are signs of deterioration (shrinking, cracking, or discoloration due to oxidation of media) or contamination. The expiration date applies to the product in its original packaging and stored as directed. Do not use product past the expiration date shown on the label.

**Shelf Life:** 90 days from date of manufacture.

## Procedure

**Specimen Collection:** Protect specimens for anaerobic culture from oxygen during collection, transportation, and processing. Consult appropriate references for detailed instructions concerning collection and transportation of anaerobes.

**Methods for Use:** BSM agar should be inoculated directly with clinical specimen or from a broth that has been inoculated with a clinical specimen. Streak plates with inoculum to obtain isolated colonies and immediately place into an anaerobic atmosphere, incubating at 35-37°C for 18-48 hours. Extended periods of incubation may be required to recover some anaerobes. Extended incubation time may also result in loss of selectivity of the media which can result in the overgrowth of organisms that should be inhibited. Detailed instructions for processing anaerobic cultures can be found in the listed references.

## Materials Required But Not Provided

Standard microbiological supplies and equipment such as loops, saline blanks, slides, staining supplies, microscope, incinerator / autoclave, incubators, anaerobic chamber / anaerobic jars, disinfectant, other culture media, and serological / biochemical reagents.

## Interpretation of Results

BSM supports good growth of *Clostridium botulinum* from food or clinical infections. This medium supports the typical lecithinase and lipase reactions produced by some Clostridia species. Degradation of lecithin in the egg yolk, from lecithinase positive organisms, results in an opaque precipitate surrounding the colonies. Destruction of the fats within the egg yolk, from lipase positive organisms, results in an iridescent sheen on the colony surface.

## Limitations

BSM will not provide complete information for identification of bacterial isolates. Additional test procedures and media are required for complete identification. It is recommended that a non-selective media, such as Brucella Blood Agar (BRU, Cat. #: AS-111) also be inoculated from the same clinical specimen to assure recovery of all species present. Consult reference materials for additional information.

## Quality Control

The following organisms are routinely used for quality assurance testing at Anaerobe Systems.

| Organism Tested               | ATCC # | Results   | Time          | Special Reaction     |
|-------------------------------|--------|-----------|---------------|----------------------|
| Bacteroides fragilis          | 25285  | No Growth |               |                      |
| Prevotella melaninogenica     | 25845  | Inhibited |               |                      |
| Fusobacterium necrophorum     | 25286  | No Growth |               |                      |
| Fusobacterium nucleatum       | 25586  | No Growth |               |                      |
| Clostridium perfringens       | 13124  | Inhibited |               | Lecithinase reaction |
| Clostridium difficile         | 9689   | Inhibited |               |                      |
| Clostridium novyi             | 7659   | No Growth |               |                      |
| Clostridium sporogenes        | 3584   | Growth    | 24 – 48 hours | Lipase reaction      |
| Propionibacterium acnes       | 6919   | No Growth |               |                      |
| Peptostreptococcus anaerobius | 28337  | No Growth |               |                      |
| Escherichia coli              | 25922  | No Growth |               |                      |
| Staphylococcus aureus         | 25923  | No Growth |               |                      |

**User Quality Control:** The final determination to the extent and quantity of user laboratory quality control must be determined by the end user.

If sterility testing is to be performed on this product, a representative sample of the lot(s) should be incubated anaerobically and aerobically for 48 – 96 hours.

If the nutritive/inhibitory capacity of this medium is to be tested for performance, it is recommended that the following ATCC organisms be evaluated for growth/inhibition.

| Organism       | ATCC # | Expected Growth | Special Reaction |             |
|----------------|--------|-----------------|------------------|-------------|
|                |        |                 | Lipase           | Lecithinase |
| B. fragilis    | 25285  | No Growth       | -                | -           |
| F. necrophorum | 25286  | No Growth       | -                | -           |
| C. sporogenes  | 3584   | 24 hours        | +                | -           |
| E. coli        | 25922  | No Growth       | -                | -           |

**Physical Appearance:** BSM should appear opaque and pale yellow in color.

## References

1. Dowell, V. R., Jr., G. L. Lombard, F. S. Thompson and A. Y. Armfield. 1977. *Media for the Isolation, Characterization and Identification of Obligately Anaerobic Bacteria*. USDHHS, CDC. Atlanta, GA 30333.
2. Engelkirk, P. G., Duben-Engelkirk, J. and Dowell, V. R. 1992. *Principles and Practices of Clinical Anaerobic Bacteriology*. Star Publishing Co., Belmont, CA 94002.
3. Holdeman, L. V., F. P. Cato and W. E. C. Moore. 1987. *Anaerobe Laboratory Manual*. Virginia Polytechnic Institute and State University. Blacksburg, VA 24061
4. Jousimeis-Somer, H. R., Summanen, P., Citron, D. M., Baron, E. J., Wexler, H. M. and S. M. Finegold. 2002. *Wadsworth – KYL Anaerobic Bacteriology Manual*. Star Publishing Co., Belmont, CA 94002.
5. CLSI. *Quality Control for Commercially Prepared Microbiological Culture Media; Approved Standard- Third Edition*. (2004). CLSI document M22-A3. CLSI, 940 West Valley Road, Suite 1400, Wayne, PA 19087-1898.
6. Dezfulian, M., McCroskey, L. M., Hatheway, C. L., & Dowell, V. R. (1981). Selective medium for isolation of Clostridium botulinum from human feces. *Journal of Clinical Microbiology*, 13(3), 526–531.

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