

# **Technical Information**

# SPS Agar, Modified

### Product Code: DM 1898

Application: - SPS Agar, Modified is used for the selective isolation and enumeration of *Clostridium perfringens* from foods.

Composition**		
Ingredients	Gms / Litre	
Casein enzymic hydrolysate	15.000	
Yeast extract	10.000	
Ferric citrate	0.500	
Sodium sulphite	0.500	
Sodium thioglycollate	0.100	
Polysorbate 80	0.050	
Sulphadiazine	0.120	
Polymyxin B sulphate	0.010	
Agar	15.000	
Final pH ( at 25°C)	7.0±0.2	
**Formula adjusted, standardized to suit performan	ce parameters	

### Principle & Interpretation

Based on the Wilson and Blair medium and the medium described by Mossel et al <sup>(2, 3)</sup> for selective isolation and enumeration of *Clostridium perfringens* from foods. SPS (Sulphite Polymyxin Sulphadiazine) Agar was developed by Angelotti et al <sup>(1)</sup> The modified SPS Agar however obviates the inclusion of Miller-Prickett tubes one of the requisite in the medium of Mossel et al <sup>(4)</sup> Casein enzymic hydrolysate and yeast extract supplies nitrogenous compounds, vitamin B complex and other essential growth nutrients to the growing *perfringens* This organism reduces sulphite to sulphide which reacts with iron of ferric *Clostridium* citrate to form a black precipitate of iron sulphide and hence the colonies are black <sup>(4)</sup>. Polysorbate 80 monooleate supplies fatty acids for the organisms. Polymyxin and Sulphadiazine inhibit a wide variety of gram-positive and gram-negative bacteria <sup>(5)</sup>. Few organisms found in food other than *Clostridium perfringens* also form black colonies on this medium. Some strains of *Clostridium perfringens* fail to grow on this medium.

# Methodology

Suspend 41.28 grams of powder media in 1000 ml distilled water. Shake well and heat to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 50°C and pour in sterile Petri plates containing inoculum. Allow to solidify and if desired, pour the cover layer using about 5 ml sterile medium. Incubate anaerobically.

## **Quality Control**

#### Physical Appearance

Cream to beige homogeneous free flowing powder

**Gelling** Firm, comparable with 1.5% Agar gel

**Colour and Clarity of prepared medium** Medium amber coloured slightly opalescent gel forms in Petri plates





Bases / Media Supplements

Reaction

Reaction of 4.13% w/v aqueous solution at 25°C. pH : 7.0±0.2

#### **pH Range** 6.80-7.20

#### Cultural Response/Characteristics

DM 1898: Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours under anaerobic conditions.

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony
Clostridium perfringens ATCC 13124	50-100	Good-luxuriant	>=50%	Black
Clostridium sporogenes ATCC 11437	50-100	Fair-good	30-40%	Black
Staphylococcus aureus ATCC 25923	50-100	None-poor	<=10%	
Escherichia coli ATCC 25922	>=10 <sup>3</sup>	inhibited	0%	white

### Storage and Shelf Life

Dried Media: Store below 30°C in tightly closed container and use before expiry date as mentioned on the label. Prepared Media: 2-8<sup>0</sup> in sealable plastic bags for 2-5 days.

# Further Reading

1 . Angelotti R., Han H. E., Foter M. J. and Lewis K. H., 1962, Appl. Microbiol., 10:193.

2. Mossel D. A. A., De Bruit A. S., Van Dipen H. M. J., Vendring

3. C. M. A. and Zoutewelle G., 1956, J. Appl. Microbiol., 19:142.

4. Mossel R. S., 1959, J. Sci. Food Agric., 19:662.

5. Downes F. P. and Ito K., (Eds.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th Ed., APHA, Washington, D.C.

6. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore.

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