SPS Agar, Modified

M898

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 performance parameters

Directions

Suspend 41.28 grams in 1000 ml distilled water. Heat to boiling 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.

Principle And Interpretation

SPS (Sulphite Polymyxin Sulphadiazine) Agar was developed by Angelotti et al (1) based on the Wilson and Blair medium and the medium described by Mossel et al (2, 3) for selective isolation and enumeration of *Clostridium perfringens* from foods. The medium of Mossel et al included the use of Miller-Prickett tubes. The modified SPS Agar however obviates the inclusion of Miller-Prickett tubes.

Casein enzymic hydrolysate and yeast extract supplies nitrogenous compounds, vitamin B complex and other essential growth nutrients to the growing *Clostridium perfringens* This organism reduces sulphite to sulphide which reacts with iron of ferric citrate to form a black precipitate of iron sulphide and hence the colonies are black (4). Polysorbate 80 monooleate supplies fatty acids for the organisms. Polymyxin and Sulphadiazine inhibit a wide variety of gram-positive and gram-negative bacteria (5). 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.

Quality Control

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

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

pH
6.80-7.20

Cultural Response
M898: 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%	white
Escherichia coli ATCC 25922	>=10^3	inhibited	0%	

Storage and Shelf Life

Store dehydrated medium and prepared medium at 2 - 8°C. Use before expiry date on the label.

Reference

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.

Revision : 02 / 2015