



## Mitis Salivarius Agar Base

M259

Mitis Salivarius Agar is recommended for the isolation of streptococci, especially *Streptococcus mitis*, *Streptococcus salivarius* and *Enterococcus faecalis* from grossly contaminated specimens.

### Composition\*\*

Ingredients	Gms / Litre
Casein enzymic hydrolysate	15.000
Peptic digest of animal tissue	5.000
Dextrose	1.000
Sucrose	50.000
Dipotassium phosphate	4.000
Trypan blue	0.075
Crystal violet	0.0008
Agar	15.000
Final pH ( at 25°C)	7.0±0.2

\*\*Formula adjusted, standardized to suit performance parameters

### Directions

Suspend 90.07 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Dispense and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 50-55°C and add 1 ml of sterile 1% Potassium Tellurite Solution (FD052). Do not reheat the medium after the addition of tellurite solution. Mix well and pour into sterile Petri plates.

### Principle And Interpretation

*Streptococcus* species are mostly commensal residents of the mouth and throat, though several may act as opportunistic pathogens and a few as primary pathogens (1). *Streptococcus* “viridans” group consists of *Streptococcus salivarius* and *Streptococcus mitis*. They exhibit different types of haemolysis when grown on Blood Agar Base. Therefore it is difficult to differentiate these organisms found in saliva from the other accompanying flora. Mitis Salivarius Agar Base is used for the isolation of *S.mitis*, *S. salivarius* and *Enterococcus faecalis* from mixed cultures. *E. faecalis* is the most common member of the Enterococci to cause infections in humans and is also a cause of human endocarditis (2). Mitis Salivarius Agar is formulated as per Chapman (3-5). This medium (with 1% potassium tellurite) is a highly selective medium, which enables to isolate streptococci from highly contaminated specimens like exudates from body cavities and faeces etc., as it inhibits a wide variety of bacteria. Some authors have also used sodium azide in this medium to inhibit the growth of gram-negative bacteria like *Proteus* (6).

Casein enzymic hydrolysate and peptic digest of animal tissue in the medium provide the essential growth nutrients. Dextrose and sucrose are the fermentable carbohydrates. Dipotassium phosphate buffers the medium. Trypan blue is an acidic, blue diazo dye while crystal violet is a basic dye and also a bacteriostatic agent, which inhibits many gram-positive organisms. Potassium tellurite also helps to make the medium selective for streptococci. Occasionally *Streptococcus mutans* strains may be inhibited on Mitis Salivarius Agar Base due to the high concentration of trypan blue in the medium. Also some *S. mitis* strains may be more easily distinguished with longer incubation (7).

### Quality Control

#### Appearance

Light yellow to light blue homogeneous free flowing powder

#### Gelling

Firm, comparable with 1.5% Agar gel

#### Colour and Clarity of prepared medium

Dark blue coloured clear to slightly opalescent gel forms in Petri plates

#### Reaction

Reaction of 9.0% w/v aqueous solution at 25°C. pH : 7.0±0.2

### pH

6.80-7.20

### Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours with added 1% Potassium Tellurite (FD052).

### Cultural Response

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony
<b>Cultural Response</b> <i>Enterococcus faecalis</i> ATCC 29212	50-100	good-luxuriant	≥50%	blue - black
<i>Escherichia coli</i> ATCC 25922	≥10 <sup>3</sup>	inhibited	0%	
<i>Staphylococcus aureus</i> ATCC 25923	≥10 <sup>3</sup>	inhibited	0%	
<i>Streptococcus intermedius</i> ATCC 9895	50-100	good-luxuriant	≥50%	blue
<i>Streptococcus pyogenes</i> ATCC 19615	50-100	good-luxuriant	≥50%	blue
<i>Streptococcus salivarius</i> ATCC 13413	50-100	good-luxuriant	≥50%	blue (gum drop)

### Storage and Shelf Life

Store below 30°C in tightly closed container and the prepared medium at 2 - 8°C. Use before expirydate on the label.

### Reference

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