Mitis Salivarius Agar Base (DM168)

Intended Use

Mitis Salivarius Agar Base is recommended for isolation of *Streptococci* from mixed cultures, especially *Streptococcus mitis*, *Streptococcus salivarius* and *Streptoccus faecalis*.

Product Summary and Explanation

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*. These organisms play a role in cariogenesis and infective endocarditis, and cause an increasing number of bacteremias.[2] Enterococci cause urinary tract infections; wound infections, bacteremia, and can colonize the skin and mucous membranes.[3] 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 facilitates the isolation of *S. mitis, S. salivarius* and *Enterococcus faecalis* from mixed cultures.[4] *E. faecalis* is the most common member of the Enterococci to cause infections in humans and is also a cause of human endocarditis.[5] Mitis Salivarius Agar base is formulated as per Chapman.[6-8] This medium (with 1% potassium tellurite (MS024) 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.[9]

Principles of the Procedure

Mitis Salivarius Agar base contains casein enzymic hydrolysate and peptic digest of animal tissue sources of carbon, nitrogen, vitamins and minerals essential for growth. Dextrose and sucrose are the fermentable carbohydrates. Dipotassium phosphate buffers the medium. Trypan blue is an acidic, blue diazo dye, which is absorbed by the colonies, producing blue color. Crystal violet is a basic dye and also a bacteriostatic agent, which inhibits many gram-positive organisms. Trypan 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.

Formula / Liter

Ingredients	Gms / Liter
Casein enzymic hydrolysate	15.00
Peptic digest of animal tissue	5.00
Dextrose	1.00
Sucrose	50.00
Dipotassium phosphate	4.00
Trypan blue	0.075
Crystal violet	0.0008
Agar	15.00
Final pH: 7.0 ± 0.2 at 25°C	
Formula may be adjusted and/or supplemented as required to meet performance specifications	

Precautions

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

Directions

1. Suspend 90.07 grams of the medium in one liter of distilled water.
2. Heat if necessary, to dissolve the medium completely.
3. Autoclave at 121°C, 15 psi pressure, for 15 minutes / validated cycle.
4. Cool to 50-55°C and add 1 ml of sterile 1% Potassium Tellurite Solution (MS024)
5. Do not reheat the medium after the addition of tellurite solution.
6. Mix well and pour into sterile petri plates.

PRODUCT SPECIFICATION SHEET

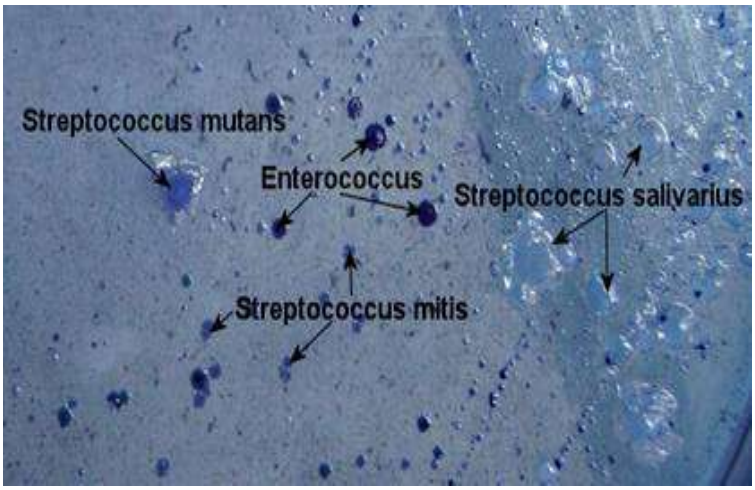
Quality Control Specifications

Dehydrated Appearance	Light yellow to light blue homogeneous free flowing powder
Prepared Medium	Dark blue coloured clear to slightly opalescent gel forms in Petri plates
Reaction of 9.0% Solution	pH : 7.0 ± 0.2 at 25°C
Gel Strength	Firm, comparable with 1.5% Agar gel

Expected Cultural Response: Cultural characteristics observed after an incubation 35 - 37°C for 18–48 hours.

Sr. No.	Organisms	Results to be achieved			
		Inoculum (CFU)	Growth	Recovery	Colour of colony
1.	*Enterococcus faecalis ATCC 29212*	50 -100	good-luxuriant	>=50 %	blue – black
2.	*Escherichia coli ATCC 25922*	>=10^3	Inhibited	0%	--
3.	*Staphylococcus aureus ATCC 25923*	>=10^3	Inhibited	0%	--
4.	*Streptococcus intermedius ATCC 9895*	50 -100	good-luxuriant	>=50 %	Blue
5.	*Streptococcus pyogenes ATCC 19615*	50 -100	good-luxuriant	>=50 %	Blue
6.	*Streptococcus salivarius ATCC 13413*	50 -100	good-luxuriant	>=50 %	Blue (gum drop)

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



Test Procedure

Refer to appropriate references for specific procedures.

Results

1. *S. mitis* produces small, flat, blue colonies. These colonies may become easier to distinguish with longer incubation.
2. *S. salivarius* produces blue, smooth or rough "gum drop" colonies, 1 - 5 mm in diameter depending on the number of colonies on the plate.
3. *Enterococcus* spp. form dark blue or black, shiny, slightly raised, 1 - 2 mm colonies.
4. *Strep mutans* will be undule-shaped colonies, with a granular frosted-glass appearance (making dextran from sugar)

Storage

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

Limitations of the Procedure

1. If coliforms grow on this medium, they produce brown colonies.
2. Molds will grow after two days incubation on this medium.
3. *Erysipelothrix rhusiopathiae* produces colorless, circular, convex colonies.
4. Beta-hemolytic streptococci produce colonies that resemble *S. mitis*.

Packaging

Product Name : Mitis Salivarius Agar Base.
Product Code : DM168
Available Pack sizes : 500gm

References

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3. Facklam, R. R., and D. F. Sahm. 1995. *Enterococcus*, *In* P. R. Murray, E. J. Baron, M. A. Pfaller, F. C. Tenover, and R. H. Yolken
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6. Chapman G. H., 1944, J. Bacteriol., 48, 113.
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8. Chapman G. H., 1947, Trans. N.Y., Acad. Sci. (Series 2), 1045.
9. Synder M. L. and Lichstein L. C., 1940, J. Infect. Dis., 67: 113.

Further Information

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DM168PI, Rev.0, 01.08.2008