



PRODUCT SPECIFICATION SHEET

Phenolphthalein Phosphate Agar (DM554)

Intended Use

Phenolphthalein Phosphate Agar (DM554) is recommended for identification of phosphatase positive *Staphylococcus aureus*.

Product Summary and Explanation

Phenolphthalein Phosphate Agar is used for the identification of phosphatase positive *Staphylococcus aureus*. Bacteria in the genus *Staphylococcus* are pathogens of man and other mammals. On the basis of their ability to clot blood plasma (the coagulase reaction) *Staphylococci* was divided into two groups. The coagulase-positive *Staphylococci* comprise of the most pathogenic species *Staphylococcus aureus*. The presence of *Staphylococci* in a lesion might first be suspected after examination of a direct gram stain. However, small numbers of bacteria in blood preclude microscopic examination and require culturing first.⁽¹⁾ Phosphatase has been implicated as a virulence factor for *Staphylococcus aureus*. The organisms produce both an acid and alkaline phosphates. Alkaline phosphates are repressed in the presence of inorganic phosphate in the medium. Phenolphthalein Phosphate Agar is used for the identification of phosphatase-positive colonies of *Staphylococcus aureus*, which is a coagulase-positive pathogenic strain.⁽²⁾

Principles of the Procedure

Phenolphthalein Phosphate Agar contains peptic digest of animal tissue and beef extract supply the nitrogenous compounds, growth factors and trace ingredients essential for the growth of *Staphylococcus aureus*. Sodium chloride maintains osmotic equilibrium of the medium. Sodium phenolphthalein phosphate serves as a substrate for the phosphatase enzyme. Phosphatase production is determined by the liberation of phenolphthalein, which is indicated by the change in colour of the medium. When alkali is added to this medium, the liberated phenolphthalein gives a bright pink-red colouration.

Formula / Liter

Ingredients	Gms / Liter
Peptic digest of animal tissue	5.00
Beef extract	3.00
Sodium chloride	5.00
Sodium phenolphthalein phosphate	0.012
Agar	15.00
Final pH: 7.4 ± 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 28.01 grams of the medium in one litre 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. Mix well and dispense as desired.





PRODUCT SPECIFICATION SHEET

Quality Control Specifications

Dehydrated Appearance	Cream to yellow homogeneous free flowing powder
Prepared Medium	Yellow coloured clear to slightly opalescent gel forms in tubes as slants
Reaction of 2.8% Solution	pH : 7.4 ± 0.2 at 25°C
Gel Strength	Firm, comparable with 1.5% Agar gel

Expected Cultural Response: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Sr. No.	Organisms	Results to be achieved		
		Inoculum (CFU)	Growth	Phosphatase
1.	<i>Escherichia coli ATCC 25922</i>	50 - 100	good-luxuriant	negative, no bright pink colour on addition of alkali
2.	<i>Staphylococcus aureus ATCC 25923</i>	50 - 100	good-luxuriant	positive, bright pink colour on addition of alkali
3.	<i>Staphylococcus epidermidis ATCC 12228</i>	50 - 100	good-luxuriant	positive, bright pink colour on addition of alkali

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

Test Procedure

Refer to appropriate references for specific procedures for identification of phosphatase positive *Staphylococcus aureus*.

Results

1. Phosphatase production is indicated by bright pink colouration on addition of alkali. The added alkali reacts with the liberated phenolphthalein which is indicated by change in colour.
2. Alternatively phosphatase production can be determined by following technique:
Technique: Grow *Staphylococci* overnight at 37°C on the medium place inverted plates and pour few drops of ammonia solution into the lid, read a positive culture whose colonies turn bright pink within a few minutes. Colour starts to fades.

Storage

Store the sealed bottle containing the dehydrated medium at 10 - 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. For identification, organisms must be in pure culture. Morphological, biochemical and/or serological tests should be performed for final identification.
2. Consult appropriate texts for detailed information and recommended procedures.





PRODUCT SPECIFICATION SHEET

Packaging

Product Name : Phenolphthalein Phosphate Agar.

Product Code : DM554

Available Pack sizes : 100gm / 500gm

References

1. Easmon C. S. F., Adlam C., 1983, Staphylococci and staphylococcal infections. Vol. 1 and 2, Academic Press, London,
2. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, BaFurther Information

For further information
please contact your local MICROMASTER Representative.



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DM554PSS,QAD/FR/024,Rev.00/01.01.2018

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