PRODUCT SPECIFICATION SHEET

Staphylococcus Agar No. 110 w/ Azide (DM615)

Intended Use

Staphylococcus Agar No. 110 w/ Azide (DM615) is recommended for selective isolation and testing of pathogenic *Staphylococci.*

Product Summary and Explanation

Stone[1] described a culture medium where staphylococci food-poisoning produced a positive gelatinase test. Chapman, Lieb, and Curcio later reported pathogenic staphylococci strains typically ferment mannitol, form pigment, and produce gelatinase.[2] Chapman suggested adding 7.5% NaCl to Phenol Red Mannitol Agar, which led to the development of Staphylococcus Agar No. 110.[3,4] The high salt concentration contributes to the selective isolation of pathogenic staphylococci. Staphylococcus Agar No. 110 is formulated as described by Chapman[4-6] for selective isolation and enumeration of from clinical as well as nonclinical specimens. Staphylococcus Agar No. 110 with azide is used for determination *Staphylococci* of coagulase positive in meat pies even in the presence of large number of *Bacillus* species.[7] This medium is recommended by APHA.[8] The addition of blood in the medium enables to study haemolytic reaction[9] and with egg yolk enables to study lecithinase production by *Staphylococcus aureus.*[10]

Principles of the Procedure

Staphylococcus Agar No. 110 w/ Azide contains casein enzymic hydrolysate and yeast extract which provide essential growth factors like vitamins, nitrogen, carbon compounds, sulphur and trace nutrients etc. making the medium nutritive to the organisms. High salt concentration makes the medium selective. The medium is differential on the basis of ability of organism to ferment mannitol, produce pigment and gelatin liquefaction. High concentration of sodium chloride inhibits many bacterial species except *Staphylococci.* Sodium azide inhibits gram-negative organisms. Mannitol fermentation can be visualized as yellow colouration by addition of a few drops of bromo thymol blue to the areas of the plates from where colonies have been removed. Gelatin is included for testing liquefaction. Gelatin liquefaction can be seen when the plates are flooded with a saturated aqueous solution of ammonium sulphate.

Formula / Liter

Ingredients	Gms / Liter
Casein enzymic hydrolysate	10.00
Yeast extract	2.50
Gelatin	30.00
Lactose	2.00
D-Mannitol	10.00
Sodium chloride	75.00
Dipotassium phosphate	5.00
Sodium azide	0.10
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.
3. Sodium azide has a tendency to form explosive metal azides with plumbing materials. It is advisable to use enough water to flush off the disposables.

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Directions

1. Suspend 149.6 grams of medium in one liter of warm distilled water.
2. Mix thoroughly. Heat to boiling to dissolve the medium completely.
3. Autoclave at 121°C, 15 psi pressure, for 15 minutes / validated cycle.
4. Resuspend the precipitate by gentle agitation to avoid bubbles and pour the plates while the medium is hot.
5. Alternatively, cool the medium to 45 - 50°C and add blood or egg yolk if desired.

Quality Control Specifications

Dehydrated Appearance	Cream to yellow homogeneous free flowing powder
Prepared Medium	Light amber coloured clear to slightly opalescent gel forms in Petri plates
Reaction of 14.96% solution	pH 7.0 ± 0.2 at 25°C
Gel Strength	Firm, comparable with 1.5% Agar gel and 3.0% gelatin gel

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

Sr. No.	Organisms	Results to be achieved					
		Inoculum (CFU)	Growth	Recovery	Mannitol Fermentation (on addition of BTB)	Pigment Production	Gelatinase production (flooding plate with standard aqueous of solution sulphate) ammonium
1.	*Staphylococcus aureus ATCC 25923*	50-100	good-luxuriant	>=50%	positive reaction	positive	positive reaction
2.	*Staphylococcus epidermidis ATCC 12228*	50-100	good-luxuriant	>=50%	variable reaction	negative	positive reaction
3.	*Enterococcus faecalis ATCC 29212*	50-100	none-poor	<=10%	slight reaction	negative	variable reaction
4.	*Escherichia coli ATCC 25922*	>=10^3	inhibited	0%			

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

Test Procedure

Refer to appropriate references for standard test procedures concerning selection and enumeration of staphylococci.

Results

Growth of pathogenic staphylococci produces colonies with yellow-orange pigment. Refer to appropriate references and test procedures for interpretation of results.

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.

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Limitations of the Procedure

1. *Enterococcus faecalis* may grow on Staphylococcus Agar No. 110 w/ Azide as tiny colonies with mannitol fermentation. Differentiate these organisms from staphylococci with the Gram stain and catalase test.
2. Suspected staphylococci must be subcultured to Nutrient Broth, Blood Agar, BHI Broth, or Tryptose Phosphate Broth for coagulase testing as the high salt content of this medium may interfere with results.
3. Pigment production is not a reliable criterion for differentiation of staphylococcal species.
4. Consult appropriate texts for detailed information and recommended procedures.

Packaging

Product Name : Staphylococcus Agar No. 110 w/ Azide

Product Code : DM615

Available Pack sizes : 500gm

References

1. Stone, R. V. 1935. A cultural method for classifying staphylococci as of the "food poisoning" type. Proc. Soc. Exptl. Biol. Med. 33:185-187.
2. Chapman, G. H., C. W. Lieb, and L. G. Curcio. 1937. Isolation and cultural differentiation of food-poisoning staphylococci. Food Research. 2:349.
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4. Chapman, G. H. 1946. A single culture medium for selective isolation of plasma-coagulating staphylococci and for improved testing of chromogensis, plasma coagulation, mannitol fermentation and the Stone reaction. J. Bacteriol. 51:409.
5. Chapman G.H., 1948, Food Res., 13:100.
6. Chapman G.H., 1952, J. Bact., 63:147.
7. Smucker S.A. and Appleman M.D., 1964, Appl. Microbiol., 12(4):355.
8. Speck M. (Ed.), 1984, Compendium of Methods for the Microbiological Examination of Foods, 2nd ed., APHA, Washington, D.C.
9. Shaffer J. C. and McDade J. J., 1962, Arch. Environ. Health, 5:547.
1. Carter C.H., 1960, J. Bact., 79:753.

Further Information

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