PRODUCT SPECIFICATION SHEET

Arginine Dihydrolase Broth (DM535)

Intended Use
Arginine Dihydrolase Broth (DM535) is used for detection of arginine dihydrolase producing microorganisms.

Product Summary and Explanation
In 1955, Moeller introduced the decarboxylase media for detecting the production of lysine and ornithine decarboxylase and arginine dihydrolase.[1] These media are a useful adjunct to other biochemical tests for the speciation and identification of the *Enterobacteriaceae* and other gram-negative bacilli.[2-4]

Principles of the Procedure
Decarboxylase Media used for the detection of arginine dihydrolase and lysine and ornithine decarboxylase was first introduced by Moeller.[1-3] Arginine Dihydrolase Broth is used for detection of arginine dihydrolase producing microorganisms. These types of media are used to differentiate bacteria on the basis of their decarboxylating activity towards the amino acids. Arginine decarboxylase enzyme is also known as Arginine dihydrolase. Arginine decarboxylase (or dihydrolase) production by various members of enteric bacteria aids in differentiating bacteria with closely related physiological characteristics.[4] Bacteria producing arginine dihydrolase enzyme decarboxylates arginine present in this medium to putrescine. The production of amine, putrescine, elevates the pH. Bromo cresol purple is the pH indicator which forms purple colour in alkaline condition. Colour change from purple to yellow and then back to purple is positive reaction.
Peptic digest of animal tissue provide the necessary nutrients to the organisms while L-arginine stimulates the arginine dihydrolase synthesis. Dipotassium phosphate buffers the medium while sodium chloride maintains the osmotic balance. In differentiation of *Enterobacteriaceae*, control tubes without arginine must be used. If the tubes give positive purple reaction the test is considered as negative.

Formula / Liter

Ingredients	Gms / Litre
Peptic digest of animal tissue	1.00
Sodium chloride	5.00
Dipotassium hydrogen phosphate	0.30
L-Arginine	10.00
Bromo cresol purple	0.016
Agar	3.00
Final pH: 6.0 ± 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 19.31 g of the medium in one liter of distilled water.
2. Heat if necessary, to dissolve the medium completely. Distribute into distribute in 13x100 mm tubes.
3. Autoclave at 115°C, 10 psi pressure, for 15 minutes.
4. Allow the tubes to cool in an upright position.

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Quality Control Specifications

Dehydrated Appearance	Light Yellow to grey colored, homogeneous, free flowing powder
Prepared Medium	Purple colored clear to slightly opalescent gel forms in tubes as butts.
Reaction of 1.93% Solution	pH 6.0 ± 0.2 at 25°C
Gel Strength	Semisolid, compared to 0.3% 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	Motility	Arginine dihydrolase
1.	*Enterobacter aerogenes ATCC 13048*	50-100	luxuriant	Positive, growth away from stabline causing turbidity	Negative reaction, yellow colour or no colour change
2.	*Klebsiella pneumoniae ATCC 13883*	50-100	luxuriant	Negative,growth along the stabline, surrounding medium remains clear	Negative reaction, yellow colour or no colour change
3.	*Proteus vulgaris ATCC 13315*	50-100	luxuriant	Positive, growth away from stabline causing turbidity	Negative reaction, yellow colour or no colour change
4.	*Salmonella Typhi ATCC 6539*	50-100	luxuriant	Positive, growth away from stabline causing turbidity	Positive reaction, purple colour
5.	*Salmonella Typhimurium ATCC 14028*	50-100	luxuriant	Positive, growth away from stabline causing turbidity	Positive reaction, purple colour
6.	*Enterobacter sakazakii ATCC 12868*	50-100	luxuriant	Positive, growth away from stabline causing turbidity	Positive reaction, purple colour

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

Test Procedure

1. Observe aseptic techniques.
2. Subculture the isolate to be tested onto a suitable medium, streaking to obtain isolated colonies, and incubate at 35 ± 2°C for 18-24 h.
3. Inoculate the broth media by transferring one or two colonies from the surface of a fresh culture with an inoculating loop or needle and mix to distribute the culture throughout the medium.
4. If desired, overlay the medium in each tube with 1- 2 mL sterile mineral oil.
5. Incubate the tubes with caps tightened at 35 ± 2°C. Examine for growth, motility and arginine dihydrolase reactions after 18-24, 48, 72 and 96 hrs before reporting as negative. The medium will become yellow initially, and then will gradually turn purple if the dihydrolase reaction occurs and elevates the pH.

Results

1. The medium becomes purple to violet if the reaction is positive (alkaline). A yellow color indicates a negative test, i.e., the organism does not produce the appropriate enzyme.
2. Consult appropriate texts for information needed to interpret the results.[7-10]

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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.[7,10,11]

Packaging
Product Name : Arginine Dihydrolase Broth
Product Code : DM535
Available Pack sizes : 100gm / 500gm
References

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		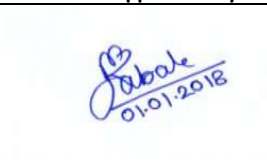
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