B1011	**DIHYDROLASE BROTH BASE**

Formula

Ingredients :	**gms/lit.**
Peptic digest of animal tissue	5.00
Yeast extract	6.00
Dextrose	2.00
Sodium chloride	30.00
Bromo cresol purple	0.032

Final pH (at 25°C) : 6.8 ± 0.2

Directions :

Suspend 43 gms.in 1000 ml. distilled water. Heat, if necessary to dissolve the medium completely. Divide in 2 parts. Add 0.5% L-arginine to first portion.Use second portion as control. Dissolve completely and dispense 3.0 ml into 13 mm X 100 mm screw cap tube. Sterilize by autoclaving at 10 lbs pressure (115 C) for 15 minutes.

Principle :

L-arginine is converted to putrescine by the dihydrolase enzyme, however the putrescine is also formed from arginine by the decarboxylase system as well. In the decarboxylase system, L-arginine undergoes decarboxylation to yield agmatine, Agmatine is then catabolized by the enzyme agmatine dehydrolase to putrescine, CO_2 and ammonia by way of an intermediate compound monocarbaminyl putrescine. It occurs in a two-step process. In the first step, hydrolytic removal of NH_2 from arginine takes place by the action of an arginine dihydrolase and rginine desimidase to yield citrullline, ammonia and inorganic phosphate. In the second step citrulline undergoes splitting or phosphorelytic cleavage by citrulline ureidase to yield ornithine and carbamylphosphate. Ornithine is then further decarboxylated to putrescine and carbon dioxide. Thus, because of production of amine like putrescine in the medium the pH is elevated. Bromo cresol purple is the pH indicator in the medium which turns purple from yellow at alkaline p. For the confirmation, it is suggested to inoculate into a basal medium tubes which does not contains L-arginine. Alkalinization of the surface of the medium may be caused by exposure to air, so a dihydrolase negative organism may be misidentified as positive. It is therefore recommende to protect the inoculated tubes from air with a layer of sterile mineral oil. Peptic digest of animal tissue and yeast extract provide nitrogenous nutrients to support bacterial growth. Dextrose is the fermentable carbohydrate.

QC Tests – (I)Dehydrated Medium	
Colour :	Light yellow
Appearance :	Homogeneous Free Flowing powder
(II)Rehydrated medium	
pH (post autoclaving/heating) :	6.8 ± 0.2
Colour (post autoclaving/heating) :	Purple
Clarity (post autoclaving/heating) :	Clear
(III)Q.C. Test Microbiological	

Cultural characteristics observed after 18 –24 hrs.at 35-37°C.

MICROORGANISM (ATCC)	GROWTH	ARGININE DIHYDROLASE
Vibrio cholerae (15748)	Luxuriant	-
Vibrio parahaemolyticus (17802)	Luxuriant	+
Enterobacter aerogenes (13048)	Luxuriant	-

Key : + = purple to yellow to purple
- = yellow

Precautions :	1. For Laboratory Use. 2. Follow proper, established laboratory procedures in handling and disposing of infectious materials.
Limitations :	1. Since the nutritional requirements of organisms vary, some strains may be encountered that fail to grow or grow poorly on this medium.
Use :	For studying dihydrolase reaction of Vibrio parahaemolyticus.
Storage :	Dehydrated medium- below 30°C Prepared medium– Between 2 to 8°C.
Packing :	500 gm bottle

Product profile:	Reconstitution	Quantity on Preparation (500g)	pH (25°C)	Supplement	Sterilization
B1011	43g/l	11.627L	6.8 ± 0.2	0.5% L-arginine	115^0C / 15 minutes