B425	**BILE ESCULIN AZIDE AGAR**
Formula	
Ingredients :	**gms/lit.**
Tryptone	17.00
Proteose peptone	3.00
Beef extract	5.00
Oxgall	10.00
Sodium chloride	5.00
Esculin	1.00
Ferric ammonium citrate	0.50
Sodium azide	0.15
Agar	15.00
Final pH (at 25°C) :	7.1 ± 0.2

Directions :

Suspend 56.65 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C. Mix well and pour into sterile Petri plates.

Principle :

This highly nutritious medium because of presence of tryptone, proteose peptone and beef extract. Sodium azide inhibits growth of gram-negative organisms and permits the cultivation of faecal Streptococci. Esculin hydrolysis and bile tolerance permit isolation and identification of group D Streptococci in 24 hours. Agar is the solidifying agent.

QC Tests – (I)Dehydrated Medium		
	Colour :	Light to medium yellow
	Appearance :	Homogeneous Free Flowing powder
(II)Rehydrated medium		
	PH (post autoclaving/heating) :	7.1 ± 0.2
	Colour (post autoclaving/heating) :	Light to medium amber
	Clarity (post autoclaving/heating) :	Clear to slightly opalescent
(III)Q.C. Test Microbiological		

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

	MICROORGANISM (ATCC)	GROWTH	ESCULIN HYDROLYSIS
	Enterococcus faecalis (29212)	Luxuriant	+
	Streptococcus bovis (27960)	Luxuriant	+
	Proteus mirabilis (25933)	Luxuriant	-
	Staphylococcus aureus (25923)	Luxuriant	-
	Streptococcus pyogenes (19615)	None - poor	-
	Key : + = blackening of medium - = no change		

Precautions :	1. For Laboratory Use.				
	2. Follow proper, established laboratory procedures in handling and disposing of infectious materials.				
	3. HARMFUL. Irritating to eyes, respiratory system and skin. Avoid contact with skin and eyes. Do not breathe dust. Wear suitable protective clothing. Keep container tightly closed. Target organ(s) : Cardiovascular, Lungs, Nerves.				
	4. 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.				
Limitations :	1. Since the nutritional requirements of organisms vary, some strains may be encountered that fail to grow or grow poorly on this medium.				
	2. Staphylococcus aureus and Staphylococcus epidermidis may exhibit growth on the medium (less than 1 mm, white – gray colonies), but they will show no action on the esculin.				
	3. Other than the enterococci, Listeria monocytogenes consistently blackens the medium around colonies. Afer 18-24 hrs., there may be a reddish to black – brown zone of hydrolysis surrounding pinpoint Listeria colonies. After 48 hours, white – gray pigmented colonies will be seen. Listeria do not attain the same degree of esculin hydrolysis displayed by enterococci in this short incubation period.				
Use :	For isolation and presumptive identification of faecal Streptococci.				
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
B425	56.65g/l	8.826L	7.1 $\pm$ 0.2	NIL	121^{0}C / 15 minutes