Bile Esculin Agar ISO

Cat #: MM-M-N031

For the isolation and presumptive identification of enterococci and for studies of fermentation of esculin by Yersinia

Principles and uses:

Bile Esculin Agar is ideal for the isolation and differentiation of intestinal enterococci, based on Esculin hydrolysis in the presence of bile. It is also reccomended by ISO 10273 for fermentation studies of esculin by Yersinia. An esculin test shall be carried out to determine presumed pathogenicity since pathogenic Yersinia enterocolitica strains are esculin negative. This test for fermentation of esculin is equivalent to the test for fermentation of salicin.
Organisms positive for esculin hydrolysis hydrolyze the glycoside esculin to esculetin and dextrose. The esculetin reacts with the Ferric citrate to form a dark brown or black colony. Bile Salts do not inhibit enterococci while other Gram positive bacteria are inhibited. Beef extract and peptone supply the nutrients essential for growth. Bacteriological agar is the solidifying agent.
Tolerance to bile and the ability to hydrolyze esculin constitutes a reliable presumptive test for the identification of Enterococci. The brown color (positive reaction) around the colonies appears after 18-24 hours of incubation at a temperature of 35±2°C.
The presence of intestinal enterococci, is an indicator for faecal contamination, especially when the contamination occurred a long before and the less resistant coliform bacteria, including Escherichia coli, may already be dead when the analysis is carried out.

Formula per Litre:

Bacteriological agar	15g	Bile salts	40g
Esculin	1g	Beef Extract	3g
Meat peptone	5g	Ferric citrate	0.5g

Preparation:

Suspend 64.5 grams of the medium in one liter of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Dispense into appropriate containers and sterilize in autoclave at 121ºC for 15 minutes. Overheating can cause darkening of the medium. If tubes are used, allow cooling in a slanted position.

Instructions for use:

For clinical diagnosis, the type of sample is bacteria isolated from feces.

- Inoculate on the surface making parallel striae with the handle or hyssop.
- Incubate in aerobic conditions at 35±2 ºC for 18-24 hours.
- Reading and interpretation of the results.

For other uses not covered by the CE marking:

Isolation and presumptive identification of enterococci:

- Streak the slant surface of the agar.
- Incubate at a temperature of 35±2ºC for 18-24 horas
- Positive cultures are confirmed on KAA Confirmatory Agar (Cat. MM-M-027) or KF Streptococcal Agar (Cat. MM-M-034).

Confirmation of pathogenic Yersinia enterocolitica according to ISO 10273:

- From the colonies selected for confirmation growth in CIN, streak the bacteria in a slanted tube of Bile Esculin Agar.
- Incubate at 30 ºC for 24±2 h.

The appearance of a black halo around the colonies indicates a positive reaction

Quality control:

Solubility	Appearance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25ºC)
w/o rests	Fine powder	Toasted	Litmus	6.6 ± 0.2

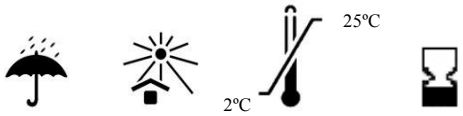
Microbiological test:

Incubation conditions: (35±2ºC / 18-28 h)
Yersinia enterocolitica according to ISO 10273 (30ºC / 24±2 h)

Microorganisms	Specification	Characteristic reaction
Streptococcus pyogenes ATCC 12344	Inhibition	
Enterococcus faecalis ATCC 19433	Good growth	Esculin Hidrolysis
Enterococcus faecium ATCC 19434	Good growth	Esculin Hidrolysis
Staphylococcus aureus ATCC 25923	Good growth	Esculin Hidrolysis (light)
Yersinia enterocolitica ATCC 27729	Good growth	
Enterococcus faecalis ATCC 29212	Good growth	Esculin Hidrolysis
Streptococcus pneumoniae ATCC 6305	Inhibition	

Storage:

Once opened keep powdered medium closed to avoid hydration.



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