

PRODUCT CODE: 415523

Bile Esculin Azide Agar (ISO 7899-2:2000) (Dehydrated Culture Media) for microbiology

Preparation

Suspend 56.6 grams of the medium in one litre 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 121°C for 15 minutes. Overheating can cause darkening of the medium. If tubes are used, allow cooling in a slanted position. The prepared medium should be stored at 8-15°C.

The colour is tournasol. The dehydrated medium should be homogeneous, free-flowing and toasted in colour. If there are any physical changes, discard the medium.

Caution: This medium is toxic if swallowed, inhaled or comes into contact with skin. Wear gloves and eye/face protection.

Uses

BILE ESCULIN AZIDE AGAR is a modification of Bile Esculin Agar with the addition of sodium azide as an inhibitor and with the reduction of the bile concentration. The resulting medium is more selective but still provides rapid growth and efficient recovery of enterococci.

The ability to hydrolyse esculin in the presence of bile is a characteristic of *enterococci*. Organisms positive for esculin hydrolysis hydrolyse the glycoside esculin to esculetin and dextrose. The esculetin reacts with the Ferric citrate to form a dark brown or black colony. Ox bile does not inhibit enterococci while other Gram-positive bacteria are inhibited. Sodium azide inhibits Gram negative bacteria. Tryptone, Peptone and Yeast extract supply the nutrients essential for growth. Sodium chloride provides the osmotic balance. Bacteriological agar is the solidifying agent.

The presence of intestinal *enterococci*, also known as faecal *streptococci*, 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. Tolerance to bile and the ability to hydrolyse esculin constitutes a reliable presumptive test for the identification of *enterococci*.

The brown colour (positive reaction) around the colonies appears after 18 - 24 hours of incubation at a temperature of $35 \pm 2^\circ\text{C}$. For the confirmation of enterococci, according to ISO 7899-2, transfer the membrane with colonies, without inverting them, to a plate with Bile Esculin Azide Agar, pre-heated to 44°C, and incubate at $44 \pm 0.5^\circ\text{C}$ for 2 hours. The plate should be read immediately.

It is considered that the typical colonies that show a brown-black colour in the surrounding medium give positive reactions and are recounted as intestinal *Enterococcus*.

Composition

See in Data Sheet (TDS).

Microbiological Test

The following results were obtained in the performance of the medium from type cultures after incubation at a temperature of 35°C ± 2°C and observed after 18-24 hours.

Microorganism	Growth	Esculin
<i>*Enterococcus faecalis</i> ATCC 11700	Good	+
<i>*Enterococcus faecium</i> ATCC 8043	Good	+
<i>Streptococcus pyogenes</i> ATCC 12344	Null	-
<i>Escherichia coli</i> ATCC 25922	Null	-
<i>Enterococcus faecium</i> ATCC 6057	Good	-

*Incubate at 44± 0.5°C for 2 hours (ISO 7899-2)

Storage

Once opened keep powdered medium closed to avoid hydration.

JM180914