

Bile Esculin Agar (i23018)

Bile Esculin Agar is used to differentiate *enterococci* and the *Streptococcus bovis* group from other *streptococci*.

Industry: Clinical / Food

Principles & Uses

Bile Esculin Agar, originally devised by Swan for the isolation and identification of group D *streptococci* from food, capitalizes on the unique ability of *enterococci* and group D *streptococci* to hydrolyze esculin, resulting in the formation of esculetin and dextrose. The interaction of esculetin with ferric citrate generates a distinct brownish-black precipitate around colonies, making it an effective indicator for their presence. Initially, this test was employed for identifying *enterococci*, but its applicability extended to group D *streptococci* as well. While the medium is highly nutritious, containing carbon sources like peptone, essential growth nutrients, and vitamins, the addition of bile ensures the inhibition of most accompanying bacteria, emphasizing its selectivity. Although Bile Esculin Agar is a valuable tool for the differentiation of *enterococci* and group D *streptococci*, other tests like salt tolerance are recommended for a comprehensive identification process.

Moreover, this medium, in accordance with ISO standards, serves as an excellent tool for fermentation studies of esculin by *Yersinia*. Pathogenic *Yersinia enterocolitica* strains are esculin negative, and this test allows for their differentiation. The organisms that display positive esculin hydrolysis can be identified by the formation of a dark brown or black colony due to their interaction with ferric citrate. Bile salts in the medium don't inhibit *enterococci*, making it a reliable presumptive test for their identification. In the context of environmental analysis, the presence of intestinal *enterococci*, indicative of fecal contamination, can be a critical measure, especially when *coliform* bacteria are no longer viable due to prior contamination. The appearance of the brown color around colonies

typically occurs after 18-24 hours of incubation at around 35°C.

Composition (gr/L)

Peptone 5, Beef Extract 3, Oxgall 20, Esculin 1, Ferric citrate 0.5, Agar 14.

Final pH at 25°C 6.8 ± 0.1

Preparation from dehydrated Powder

Suspend 43.5 g of the powder in 1 L of purified water. Autoclave at 121°C for 15 minutes.

Quality Control

Dehydrated Appearance: Fine, homogeneous, free of extraneous material, may contain a moderate amount of very small dark particles.

Prepared Appearance: Dark, tan olive to olive green with a blue tint, trace hazy to hazy. Reaction of 4.35 % Solution at 25°C: pH 6.8 ± 0.1

Cultural Response

Cultural response was observed after 42-48 hours of incubation at 35 ± 2°C.

Organism (ATCC*)	Recovery	Colony color
<i>Enterococcus faecalis</i> (29212)	Good	Black
<i>Streptococcus pyogenes</i> (19615)	Partial to complete inhibition	Colorless

*ATCC is a registered trade mark of the American Type Culture Collection.



E. faecalis exhibits a brownish medium around colonies due to esculin hydrolysis.

Storage

Keep the container at 15-30 °C. Store prepared medium at 2-8 °C.