

## TECHNICAL DATA SHEET

### Bile Esculin Agar

#### Principle

The media is formulated by Swan (1954) and modified by Facklam and Moody (1970). The media composition and performance criteria are as per the specification laid down in ISO 10273:1994. Bile esculin agar is composed of meat extract (Equivalent to beef extract), peptone, oxgall, ferric citrate, esculin and agar. Meat extract and peptone provides nitrogen and other necessary minerals. Oxgall is selective agent inhibits gram positive bacteria other than enterococci. Ferric citrate is indicator for esculin hydrolysis. The esculin (glycoside) is source of carbon. The esculin hydrolyzing microorganisms hydrolyzes esculin to esculetin and dextrose. The esculetin reacts with ferric citrate to form brown or black color complex. Agar is solidifying agent

**Use:** For the isolation and presumptive identification of group D Streptococci.

#### Contents\*

Ingredients	Gram/Litre
Meat Extract#	3.00
Peptone	5.00
Oxgall	40.00
Ferric Citrate	0.50
Esculin	1.00
Agar	15.00
pH at 25°C	6.6 ±0.2

\* Formula adjusted for optimum performance and parameters # Equivalent to Beef Extract

**Directions:** Dissolve 64.50 grams in 1000 ml distilled water. Boil to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 min (Overheating may causes darkening of medium), cool it to 42-45 °C and dispense in desired (Test tubes to form butt and in petri plates for isolation and identification). Ensure complete solidification and inoculate test sample aseptically.

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## Specimens' types analyzed Food and fooder samples etc.

### Precautions to be taken

These microbial media are intended for the in-vitro use only. All the handling, experiments, storage, and discarding should be performed with the help of skilled and knowledgeable technicians and as per the established guidelines. The material should be disposed only after proper sterilization by autoclaving. Please go through the MSDS of the media to avoid any accidents or in emergency.

### Performance and Evaluation

The expected performance of the medium is liable to use as per the direction on the label when stored at optimum conditions and within expiry date.

### Quality Control

Appearance	Light beige colored free flowing, homogeneous powder
Reaction of 6.45% solution	6.6 ±0.2 at 25 °C
pH	6.40- 6.80
Gelling	Firm comparable with 1.5% agar gel
Color and clarity of ready medium	Amber colored opalescent gel
Growth Promotion Properties	Best at ≤ 100 CFU at 32-37 °C for 18-48 h
Indicative properties	Optimum at ≤ 100 CFU at 32-37 °C for 18-24 h
Negative control	Performed using sterile distilled water

### Different Microbial Response

Cultural characteristics observed after incubation at 33-37°C for 18-24 hours. Inoculum 50-100 CFU.

Organism	ATCC	Growth	Recovery	Esculin hydrolysis
<i>Yersinia enterocolitica</i>	27729	Luxuriant	≥ 60%	Positive (Black color)
<i>Enterococcus faecalis</i>	14506	Luxuriant	≥ 60%	Positive (Black color)
<i>Escherichia coli</i>	8739	Good	≥ 50%	Negative

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**Storage and Shelf Life:** The product is highly hygroscopic; keep the container tightly closed at all times and store it properly as per the conditions mentioned on the label. The declared expiry is valid only when stored as per the conditions mentioned on the label. Note: Sterilize media immediately after reconstitution.

**Disposal:** To avoid the contamination or propagation of any hazardous microbes the used, unusable or modified preparation of this product must be disposed after autoclaving after completion of task.

## Reference

1. Atlas, R. M. (2005). *Handbook of media for environmental microbiology*. CRC press.
2. *Difco Manual* (1998). 11<sup>th</sup> Edition. Difco Laboratories., Division of Becton Dickinson and Company, Sparks, Maryland, USA.
3. Facklam, R. R., and M. D. Moody. (1970). *Presumptive identification of group D streptococci: The bile-esculin test*. Appl. Microbiol. 20:245.
4. Swan, A. (1954). *The use of bile-esculin medium and of Maxted's technique of Lancefield grouping in the identification of enterococci (group D streptococci)*. J. Clin. Pathol. 7:160.

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