EVA Broth

Intended Use

EVA (Ethyl Violet Azide) Broth is used for detecting and confirming enterococci in water and other specimens as an indication of fecal contamination.

Summary and Explanation

The presence of enterococci in water and other specimens indicates fecal contamination. Mallmann and Seligmann[1] compared various enrichment media for detecting fecal streptococci and found that Azide Dextrose Broth presumptively identified the streptococci. However, because gram-positive bacteria other than enterococci grow in that medium, confirmation is necessary. Litsky et al.[2] studied various dyes and selective agents and formulated a medium using ethyl violet and sodium azide as selective agents. The medium known as Ethyl Violet Azide (EVA) Broth is specific for enterococci. In conjunction with Azide Dextrose Broth, EVA Broth is used to confirm the presence of enterococci.

Principles of the Procedure

EVA Broth contains peptones as sources of carbon, nitrogen, vitamins and minerals. Dextrose is the carbohydrate. Sodium azide and ethyl violet inhibit gram-positive bacilli and gram-positive cocci other than enterococci. Monopotassium and dipotassium phosphates buffer the medium. Sodium chloride provides osmotic balance.

Formula

Difco™ EVA Broth

Approximate Formula* Per Liter

Proteose Peptone No. 3	8.0	g
Pancreatic Digest of Casein	12.0	g
Dextrose	5.0	g
Dipotassium Phosphate	2.7	g
Monopotassium Phosphate	2.7	g
Sodium Chloride	5.0	g
Sodium Azide	0.4	g
Ethyl Violet	0.83	mg

**Adjusted and/or supplemented as required to meet performance criteria.*

Directions for Preparation from Dehydrated Product

1. Dissolve 35.8 g of the powder in 1 L of purified water. Mix thoroughly.
2. Autoclave at 121°C for 15 minutes.
3. Test samples of the finished product for performance using stable, typical control cultures.

Procedure

See appropriate references for specific procedures.

Expected Results

Growth of enterococci.

User Quality Control

Identity Specifications

Difco™ EVA Broth

Dehydrated Appearance:	Light beige, free-flowing, homogeneous.
Solution:	3.58% solution, soluble in purified water. Solution is light amber, clear to very slightly opalescent.
Prepared Appearance:	Light amber, clear to very slightly opalescent.
Reaction of 3.58% Solution at 25°C:	pH 7.0 ± 0.2

Cultural Response

Difco™ EVA Broth

Prepare the medium per label directions. Inoculate and incubate at 35 ± 2°C for 18-48 hours.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY
Enterococcus faecalis	19433	10^2-10^3	Good
Enterococcus faecalis	29212	10^2-10^3	Good
Escherichia coli	25922	10^3	Inhibition

Uninoculated Tube — *Enterococcus faecalis* ATCC™ 29212

References

1. Mallmann and Seligmann. 1950. Am. J. Pub. Health *40*:286.
2. Litsky, Mallmann and Fifield. 1953. Am. J. Pub. Health *43*:873.

Availability

Difco™ EVA Broth

Cat. No. 212107 Dehydrated – 500 g