



Barritt Reagent B (for VP test)

R030

The reagent is used in Voges-Proskauer test for detection of acetoin production by bacterial culture.

Composition**

Ingredients

Potassium hydroxide	40.000 gm
Distilled water	100.000 ml

**Formula adjusted, standardized to suit performance parameters

Directions

Grow test culture in MR-VP Medium (M070). Add 0.2 ml (2 drops) of Reagent A and 0.2 ml (2 drops) of Reagent B (R030) for 10 ml medium. Shake tubes gently for 30 seconds to 1 minute to expose the medium to atmospheric oxygen in order to oxidize the acetoin (acetylmethylcarbinol) so as to obtain a colour reaction. Allow tube to stand at least 10 to 15 minutes.

Principle And Interpretation

VP test is helpful in identifying members of the family Enterobacteriaceae. Initially all enterics will give a positive MR reaction if tested. However, after further incubation, required by the test procedure (2-5 days), MR - positive organisms continue to produce acids, resulting in a low pH (acidic) that overcomes the phosphate buffering system and maintain an acidic environment in the medium (pH 4.2 or less). MR- negative organisms further metabolize the initial fermentation products by decarboxylation to produce neutral acetyl methylcarbinol (acetoin), which results in decreased acidity in the medium and raises the pH towards neutrality (pH 6.0 or above). In the presence of atmospheric oxygen and alkali (potassium hydroxide), the neutral end products, acetoin and 2, 3-butanediol, are oxidized to diacetyl, and α -Naphthol serves as a catalyst to produce a red colour complex.

Quality Control

Appearance

Colourless solution.

Clarity

Clear solution without any precipitate. Note : On storage of the reagent, precipitate may develop. This will not affect the performance criteria of the reagent.

Cultural Response

R030: Biochemical identification was carried out by adding Barritt Reagent (Part A) (R029) and Barritt Reagent (Part B) (R030) in 24-48 hours old cultures grown in MR-VP Medium (M070).

Organism	Growth	VP Test
<i>Enterobacter aerogenes</i> ATCC 13048	Luxuriant	Positive (Red colour formation)
<i>Escherichia coli</i> ATCC 25922	Luxuriant	Negative (No red colour formation)
<i>Klebsiella pneumoniae</i> ATCC 13883	Luxuriant	Positive (Red colour formation)

Storage and Shelf Life

Store at 10-30°C in tightly closed container. Use before expiry period on the label.

Reference

1. Color Atlas and Textbook of Diagnostic Microbiology, 4th edition, Elmer W. Koneman, Stephen D. Allen, William M. Janda, Paul C. Schreckenberger, Washington C. Winn.

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