



Proline Discs

DD052

Proline discs are used for amino acid decarboxylation test.

Directions

To determine amino acid decarboxylation, the Proline disc (DD052) is added in the Decarboxylase Broth Base, Moeller (M393) which is used as a negative control for studying decarboxylation or as a base for the addition of amino acids. The test organism is inoculated into the broth containing the Proline disc (DD052). The inoculated tubes are overlaid with sterile mineral oil and incubated at 35-37°C for up to 4 days. A purple colour indicates the decarboxylation.

Principle And Interpretation

Amino acid discs are used to differentiate the microorganisms on the basis of their ability to decarboxylate the amino acids. Proline is an essential amino acid. Moeller introduced the Decarboxylase Broth for detecting the production of decarboxylase (1). Prior to Moellers work, bacterial amino acid decarboxylases were studied by Gale (2), Gale and Epps (3). Moeller Decarboxylase Broth Base (M393) contains dextrose which is the fermentable carbohydrate and pyridoxal is the co-factor for the decarboxylase enzyme. Bromo cresol purple and cresol red are the pH indicators in this medium. When the medium is inoculated with the dextrose fermenting bacteria, the pH is lowered due to acid production, which changes the colour of the indicator from purple to yellow. Acid produced stimulates decarboxylase enzyme. Amino acid decarboxylation yields amine. Formation of the amine increases the pH of the medium, changing the colour of the indicator from yellow to purple. If the organisms do not produce the appropriate enzyme, the medium remains acidic, yellow in colour. Inoculated tubes must be protected from air with a layer of sterile mineral oil. Exposure to air may cause alkalinization at the surface of the medium which makes the test invalid.

Positive Test: Colour of the medium changes from yellow to purple

Negative Test: Colour of the medium changes to yellow or there is no change

Quality Control

Appearance

Filter paper discs of 10 mm diameter

Cultural Response

Cultural characteristics observed in Moeller Decarboxylase Broth Base (M393) with added Proline discs (DD052) after incubation at 35-37°C up to 4 days (Inoculated tubes are overlaid with sterile mineral oil).

Cultural Response

Organism	Inoculum (CFU)	Proline decarboxylation
Cultural Response		
<i>Citrobacter freundii</i> ATCC 8090	50-100	variable reaction
<i>Enterobacter aerogenes</i> ATCC 13048	50-100	positive reaction, purple colour
<i>Escherichia coli</i> ATCC 25922	50-100	variable reaction
<i>Klebsiella pneumoniae</i> ATCC 13883	50-100	negative reaction, yellow colour
<i>Proteus mirabilis</i> ATCC 25933	50-100	positive reaction, purple colour

<i>Proteus vulgaris</i> ATCC 13315	50-100	negative reaction, yellow colour
<i>Pseudomonas aeruginosa</i> ATCC 9027	50-100	negative reaction, yellow colour
<i>Salmonella Paratyphi A</i> ATCC 9150	50-100	positive reaction, purple colour
<i>Salmonella Typhi</i> ATCC 6539	50-100	negative reaction, yellow colour
<i>Serratia marcescens</i> ATCC 8100	50-100	positive reaction, purple colour
<i>Shigella dysenteriae</i> ATCC 13313	50-100	negative reaction, yellow colour
<i>Shigella flexneri</i> ATCC 12022	50-100	negative reaction, yellow colour
<i>Shigella sonnei</i> ATCC 25931	50-100	positive reaction, purple colour

Storage and Shelf Life

Store the discs at 10-30°C. Use before expiry date on the label.

Reference

1. Moeller V., 1955, Acta Pathol. Microbiol. Scand. 36:158.
2. Gale G. F., 1940, Biochem. J., 34:392.
3. Gale and Epps, 1943, Nature, 152:327.

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