

# PSEUDOMONAS P AGAR

## INTENDED USE

Remel Pseudomonas P Agar is a solid medium recommended for use in qualitative procedures for differentiation of *Pseudomonas aeruginosa* from other *Pseudomonas* spp. based on pyocyanin production.

## SUMMARY AND EXPLANATION

In 1954, King et al. developed two media formulations (Medium A and Medium B) to enhance pigment production in pseudomonads.<sup>1</sup> Previous work had demonstrated amino acids, inorganic ions, minerals, and peptones influenced the production of pyocyanin and fluorescein. King determined the production of these pigments was affected by the composition of the peptone in the medium. Pigment production was also influenced by the absence or minimal concentrations of components which may have a detrimental effect. A combination of magnesium chloride and potassium sulfate has been found to most effectively stimulate the production of pyocyanin.<sup>2</sup>

## PRINCIPLE

*P. aeruginosa* is the only species of bacteria known to produce pyocyanin, a blue-green pigment which diffuses into the agar surrounding the growth. Gelatin and meat peptones provide the nutrients necessary for bacterial growth. These peptones contain less than 0.1% phosphorous which has been found to increase pyocyanin production. Magnesium chloride and potassium sulfate in a total amount that does not exceed 2% stimulates pyocyanin production. Glycerol, added to the prepared medium, is an energy source.

## REAGENTS (CLASSICAL FORMULA)\*

Gelatin Peptone .....	10.0 g	Magnesium Chloride .....	1.4 g
Meat Peptone.....	10.0 g	Agar.....	15.0 g
Potassium Sulfate .....	10.0 g	Deminerlized Water.....	1000.0 ml

pH 7.2 ± 0.2 @ 25°C

\*Adjusted as required to meet performance standards.

## PRECAUTIONS

This product is For Laboratory Use only. It is not intended for use in the diagnosis of disease or other conditions.

## PREPARATION OF DEHYDRATED CULTURE MEDIUM

1. Suspend 46.4 g of medium in 1000 ml of demineralized water containing 10 ml of glycerol.
2. Heat to boiling with agitation to completely dissolve.
3. Sterilize by autoclaving at 121°C for 15 minutes or following established laboratory procedures.
4. Dispense into appropriate containers.

## PROCEDURE

1. Consult current editions of appropriate references for the recommended procedure for sample preparation, inoculation, testing, and interpretation.

## INTERPRETATION OF THE TEST

Positive Test - Blue-green pigment diffusing into the agar surrounding growth

Negative Test - No blue-green pigment produced

## QUALITY CONTROL

Each lot number of Pseudomonas P Agar has been manufactured, packaged, and processed in accordance with current Good Manufacturing Practice regulations. All lot numbers have been tested using the following quality control organisms and have been found to be acceptable. Testing of control organisms should be performed in accordance with established laboratory quality control procedures. If aberrant quality control results are noted, sample results should not be reported.

## CONTROL

*Pseudomonas aeruginosa* ATCC® 27853

*Stenotrophomonas maltophilia* ATCC® 13637

## INCUBATION

Ambient, up to 48 h @ 33-37°C

Ambient, up to 48 h @ 33-37°C

## RESULTS

Growth, blue-green pigment

Growth, no pigment

## LIMITATIONS

1. Occasional strains of *P. aeruginosa* may fail to produce pyocyanin.<sup>3</sup>
2. Some strains of *P. aeruginosa* may produce brown-black pyomelanin or red pyorubin which may mask pyocyanin production on Pseudomonas P Agar, resulting in a variety of hues from blue to red.<sup>2</sup>
3. Pyocyanin is soluble in chloroform. If pigment production is questionable (small amount of blue-green color) confirm the reaction by extraction with chloroform. Add several drops of chloroform to the growth and observe for a blue-green color.<sup>2</sup>

## BIBLIOGRAPHY

1. King, E.O., M.K. Ward, and D.E. Raney. 1954. J. Lab. Clin. Med. 44:301-307.
2. MacFaddin, J.F. 1985. Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria. Vol. 1. Williams & Wilkins, Baltimore, MD.
3. Gaby, W.L. and E. Free. 1953. J. Bacteriol. 65:746.

Refer to the front of Remel *Technical Manual of Microbiological Media* for **General Information** regarding precautions, product storage and deterioration, sample collection, storage and transportation, materials required, quality control, and limitations.

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