Iron Oxidizing Medium (Twin Pack)

M615

Iron Oxidizing Medium is used for the isolation, cultivation and enrichment of *Thiobacillus ferroxidans* .

Composition**

Ingredients	Gms / Litre
Part A	-
Ammonium sulphate	3.000
Potassium chloride	0.100
Dipotassium phosphate	0.500
Magnesium sulphate. heptahydrate	0.500
Calcium nitrate	0.010
Part B	-
Ferrous sulphate. heptahydrate	44.220
Final pH (at 25°C)	3.3±0.3

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 3.85 grams (the equivalent weight of dehydrated medium per litre) of Part A in 700 ml distilled water containing 1 ml of 10N sulphuric acid. Heat to boiling to dissolve the medium completely. Suspend 24.16 grams (the equivalent weight of dehydrated medium per litre) of Part B separately in 300 ml distilled water. Heat if necessary to dissolve the medium completely. Sterilize Part A and Part B separately by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool each solution to 25°C. Aseptically add 300 ml of sterilized Part B to 700 ml of Part A. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Principle And Interpretation

Thiobacillus ferrooxidans is recognized as being responsible for the oxidation of iron and inorganic sulfur compounds in areas such a mine tailings and coal deposits where these compounds are abundant (1,2). The main importance of *T. ferrooxidans* has been in acid mine drainage. T. ferrooxidans is generally assumed to be obligately aerobic, but under anaerobic conditions, *T.ferrooxidans* can be grown on elemental sulfur using ferric iron as an electron acceptor. These results indicate that *T.ferrooxidans* can be considered as facultative anaerobe playing an important role in the iron and sulfur cycles in acidic environments. The ability of this organism to grow in oxygen-deficient environments may have important implications in bioleaching processes where anaerobic conditions may often exist (3). Iron Oxidizing Medium (! Thiobacillus ferroxidans) is formulated in accordance with APHA (4) and is used for isolation, cultivation and enrichment of *T.ferroxidans* .

Magnesium sulphate, ammonium sulphate, potassium chloride and calcium nitrate are sources of ions that stimulate metabolism. Dipotassium phosphate buffers the medium. The medium has a precipitate, is opalescent and green in colour.

T.ferroxidans utilizes ferrous sulphate as energy source. Some oxidation of iron occurs during sterilization. *T. ferroxidans* can be enumerated by MPN technique (5). Growth of the organism is manifested by a decrease in pH and an increase in concentration of oxidized iron. With the use of uninoculated controls, an increase of deep orange brown colour can be seen in positive enrichment tubes or flasks as compared to negative ones. The organisms are highly / strictly aerobic, so the tubes should be shaken every day during incubation.

Quality Control

Appearance of Part A

White to cream homogeneous free flowing powder.

Appearance of Part B

Greenish yellow to dark green Homogeneous hygroscopic Powder

Colour and Clarity of Prepared medium

Brownish yellow clear to slightly opalescent with precipitate.

Reaction
Reaction of (0.39 gm Part A in70 ml distilled water containing 1 ml of 10N sulphuric acid)+ 2.42 gm of Part B in 30 ml distilled water at 25°C. pH : 3.3±0.3

pH

3.00-3.60

Cultural Response
Cultural characteristics observed after an incubation at 30°C upto 5 days.

Cultural Response

Organism	**Growth**
Cultural Response	
Thiobacillus ferrooxidans *ATCC 23270*	luxuriant

Storage and Shelf Life
Store below 30°C in tightly closed container and prepared medium at 2-8°C. Use before expiry period on the label.

Reference
1.Unz R. F. and Lundgren D. G., 1961, Soil Sci., 92:302.
2.McGoran C. J .M., Duncan D. W. and Walden C. C., 1969, Can. J.Microbiol., 15:135.

3.Pronk T. T., de Bruyn J. C., Bos P. and Kuenen J. G., 1994Appl. Environ. Microbiol., 58. 2227-2230.

4.Eaton A. D., Clesceri L. S., Rice E. W. and Greenberg A. W.(Eds.), 2005, Standard Methods for the Examination of Water and Wastewater, 21st Ed., APHA, Washington, D.C.

5.Silverman M. P. and Lundgren D. C., 1959, J. Bacteriol 77:642.

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