Technical Information

Iron Oxidizing Medium (Twin Pack)

Product Code: DM 1615

Application: - 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	44.220
Final pH (at 25°C)	3.3±0.2

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

Thiobacillus ferrooxidans is famous 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 supposed 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 feature in bioleaching processes where anaerobic conditions may often exist [3]. Iron Oxidizing Medium (! Thiobacillus ferroxidans) is formulated as per the APHA [4] guidance 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 is opalescent and green in colour having a precipitate

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.

Methodology

Suspend 3.85 grams of dehydrated medium Part A in of powder media 700 ml distilled water containing 1 ml of 10 N sulphuric acids. Shake well & heat if necessary to dissolve the medium completely. Suspend 44.22 grams 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 and aseptically mix Part A and Part B solutions. Aseptically distribute into sterile tubes or flasks.

Quality Control

Physical Appearance

Part A :White to cream homogeneous free flowing powder Part B :Greenish yellow to dark green hygroscopic powder.

Colour and Clarity of prepared medium

Green coloured, clear solution with precipitate.

Reaction

Reaction of Part A(0.38 gm in 70 ml distilled water containing 0.1 ml of 10 N sulphuric acid)+ Part B(4.42 gm in 30ml distilled water) at 25°C. pH : 3.3±0.2

pH range

3. 10-3.50

Cultural Response/Characteristics

DM 1615: Cultural characteristics observed after an incubation at 30°C upto 5 days.

Organism	Growth
Thiobacillus ferrooxidans ATCC 23270	luxuriant

Storage and Shelf Life

Dried Media: Store below 30°C in tightly closed container and use before expiry date as mentioned on the label.
Prepared Media: 2-8^0 in sealable plastic bags for 2-5 days.

Further Reading

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., 1994,Appl. 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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