

### **ATCC medium: 1768 *Geobacter metallireducens* Medium**

Ferric citrate (Sigma F-6129).....	13.7 g
Wolfe's Vitamin Solution (see below).....	10.0 ml
Wolfe's Mineral Solution (see below).....	10.0 ml
NaHCO <sub>3</sub> .....	2.5 g
NH <sub>4</sub> Cl .....	0.25 g
NaH <sub>2</sub> PO <sub>4</sub> . H <sub>2</sub> O.....	0.6 g
KCl.....	0.1 g
Sodium acetate.....	6.8 g
Distilled, deionized water.....	1.0 L

Heat about 400 ml of water on a hot stir plate to near boiling. Add ferric citrate, allow it to dissolve, then cool the solution to room temperature in a slurry of ice. Add 400 ml of water; this quickly cools the medium to room temperature. Adjust the pH to 6.0 using 10 N NaOH; when the pH approaches 5.0, add NaOH by drops until the pH level is established. Approximately 6 ml of 10 N NaOH per liter will be needed per liter of medium. Add the remaining ingredients and bring the final volume up to 1.0 L with water. Bubble the medium with 80% N<sub>2</sub> and 20% CO<sub>2</sub>. The final pH should be 6.8-7.0.

**Do not expose this medium to sunlight.**

#### *Wolfe's Vitamin Solution:*

Available from ATCC as a sterile ready-to-use liquid (Vitamin Supplement, catalog no. MD-VS).

Biotin.....	2.0 mg
Folic acid.....	2.0 mg
Pyridoxine hydrochloride.....	10.0 mg
Thiamine . HCl.....	5.0 mg
Riboflavin.....	5.0 mg
Nicotinic acid.....	5.0 mg
Calcium D-(+)-pantothenate.....	5.0 mg
Vitamin B12.....	0.1 mg
p-Aminobenzoic acid.....	5.0 mg
Thioctic acid.....	5.0 mg
Distilled water.....	1.0 L

#### *Wolfe's Mineral Solution:*

Available from ATCC as a sterile ready-to-use liquid (Trace Mineral Supplement, catalog no. MD-TMS.)

Nitrilotriacetic acid.....	1.5 g
MgSO <sub>4</sub> . 7H <sub>2</sub> O .....	3.0 g
MnSO <sub>4</sub> . H <sub>2</sub> O .....	0.5 g
NaCl.....	1.0 g
FeSO <sub>4</sub> . 7H <sub>2</sub> O .....	0.1 g
CoCl <sub>2</sub> . 6H <sub>2</sub> O .....	0.1 g
CaCl <sub>2</sub> .....	0.1 g
ZnSO <sub>4</sub> . 7H <sub>2</sub> O .....	0.1 g
CuSO <sub>4</sub> . 5H <sub>2</sub> O .....	0.01 g
AlK(SO <sub>4</sub> ) <sub>2</sub> . 12H <sub>2</sub> O.....	0.01 g
H <sub>3</sub> BO <sub>3</sub> .....	0.01 g
Na <sub>2</sub> MoO <sub>4</sub> . 2H <sub>2</sub> O.....	0.01 g
Distilled water.....	1.0 L

Add nitrilotriacetic acid to approximately 500 ml of water and adjust to pH 6.5 with KOH to dissolve the compound. Bring volume to 1.0 L with remaining water and add remaining compounds one at a time.