

**ATCC medium: 2055 *Pyrodictium abyssi* medium**

NaCl.....	13.85 g
MgSO <sub>4</sub> . 7H <sub>2</sub> O .....	3.5 g
MgCl <sub>2</sub> . 6H <sub>2</sub> O .....	2.75 g
CaCl <sub>2</sub> . 2H <sub>2</sub> O .....	0.75 g
KH <sub>2</sub> PO <sub>4</sub> .....	0.5 g
KCl.....	325.0 mg
NaBr.....	50.0 mg
H <sub>3</sub> BO <sub>3</sub> .....	15.0 mg
(NH <sub>4</sub> ) <sub>2</sub> SO <sub>4</sub> .....	10.0 mg
SrCl <sub>2</sub> . 6H <sub>2</sub> O .....	7.5 mg
NiCl <sub>2</sub> . 6H <sub>2</sub> O .....	2.0 mg
Na <sub>2</sub> WO <sub>4</sub> . 2H <sub>2</sub> O.....	0.1 mg
KI.....	50.0 mcg
Wolfe's Mineral Solution (see below).....	10.0 ml
Yeast extract.....	0.5 g
Resazurin.....	1.0 mg
Sulfur.....	30.0 g
Na <sub>2</sub> S . 9H <sub>2</sub> O .....	0.5 g
Distilled water.....	1.0 L

Place approximately 0.3 g sulfur per 18 X 150 mm Balch tube. Mix and dissolve ingredients through resazurin in distilled water. Adjust to pH 5.5 with H<sub>2</sub>SO<sub>4</sub>. Boil and cool under 80% N<sub>2</sub>, 20% CO<sub>2</sub>. Add sodium sulfide and readjust to pH 5.5. Using the same gas phase, anaerobically dispense 10 ml medium per Balch tube containing sulfur. Crimp stoppers down tightly. Sterilize medium by tyndallization (i.e., heat tubes to 100C in boiling water for 1 hour on each of 3 consecutive days).

Inoculate tubes and overpressure to 200-300 kPa with 80% H<sub>2</sub>, 20% CO<sub>2</sub>.

*Wolfe's Mineral Solution:*

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

Nitriilotriacetic 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 nitriilotriacetic 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.