

ATCC medium: 2114 *Thermotoga* medium

Soluble starch.....	5.0 g
KH ₂ PO ₄	0.5 g
NiCl ₂ . 6H ₂ O	2.0 mg
NaCl.....	20.0 g
Yeast extract.....	0.5 g
Resazurin.....	1.0 mg
Wolfe's Mineral Solution (See below)....	15.0 ml
Na ₂ S . 9H ₂ O	0.5 g
Artificial seawater.....	250.0 ml
Distilled water.....	750.0 ml

Combine all ingredients except for Na₂S. Boil for 1-2 minutes. Allow to cool under N₂ gas. Add Na₂S. Adjust pH to 6.5. Dispense under same gas.

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 ₄ . 7H ₂ O	3.0 g
MnSO ₄ . H ₂ O	0.5 g
NaCl.....	1.0 g
FeSO ₄ . 7H ₂ O	0.1 g
CoCl ₂ . 6H ₂ O	0.1 g
CaCl ₂	0.1 g
ZnSO ₄ . 7H ₂ O	0.1 g
CuSO ₄ . 5H ₂ O	0.01 g
AlK(SO ₄) ₂ . 12H ₂ O.....	0.01 g
H ₃ BO ₃	0.01 g
Na ₂ MoO ₄ . 2H ₂ 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.