

ATCC medium: 1852 *Pelobacter* medium with gallic acid

Solution A:

NaCl.....20.0 g
MgCl₂ . 6H₂O3.65 g
KH₂PO₄0.2 g
NH₄Cl0.25 g
KCl.....0.5 g
CaCl₂ . 2H₂O0.15 g
Modified Wolfe's Minerals (see below)...10.0 ml
Wolfe's Vitamin Solution (see below)....10.0 ml
NaHCO₃.....2.5 g
Resazurin.....0.5 mg
Distilled water.....945.0 ml

Add all components except sodium bicarbonate to distilled water; heat to boiling and cool under 80% N₂, 20% CO₂. Add sodium bicarbonate and equilibrate under same gas phase. Tube anaerobically under same gas phase. Tube anaerobically under same gas phase and autoclave at 121C for 15 minutes. Add Solutions B and C prior to inoculation. Final pH of complete medium is 7.2.

Solution B:

Gallic acid.....0.85 g
Distilled water.....25.0 ml

Prepare gallic acid solution just before use. Neutralize quickly to approximately pH 6.5 and filter-sterilize. Equilibrate under 100% N₂ before adding to bulk of medium.

Solution C:

Na₂S . 9H₂O0.4 g
Distilled water.....10.0 ml

Equilibrate sodium sulfide under 100% N₂ and tube under the same gas phase. Autoclave at 121C for 15 minutes.

Modified Wolfe's Minerals:

Na₂SeO₃10.0 mg
NiCl₂ . 6H₂O10.0 mg
Na₂WO₄ . 2H₂O.....10.0 mg
Wolfe's Mineral Solution (see below).....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 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.

Wolfe's Vitamins:

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

Folic acid.....	2.0 mg
Pyridoxine hydrochloride....	10.0 mg
Riboflavin.....	5.0 mg
Biotin.....	2.0 mg
Thiamine.....	5.0 mg
Nicotinic acid.....	5.0 mg
Pantothenic acid.....	5.0 mg
Vitamin B12.....	0.1 mg
p-Aminobenzoic acid.....	5.0 mg
Thioctic acid.....	5.0 mg
Distilled water.....	1.0 L