

ATCC Medium: 1439 Methanogenium Medium

NaCl.....	18.0 g
MgCl ₂ x 7H ₂ O.....	2.75 g
KCl.....	0.335 g
MgSO ₄ x 7H ₂ O.....	3.45 g
NH ₄ Cl.....	0.25 g
CaCl ₂ x 2H ₂ O.....	0.14 g
K ₂ HPO ₄	0.14 g
Trace Elements (see below).....	10.0 ml
Vitamin Solution (see below).....	10.0 ml
Fe(NH ₄) ₂ (SO ₄) ₂ x 7H ₂ O.....	2.0 mg
NaHCO ₃	5.0 g
Na Acetate.....	1.0 g
Yeast Extract.....	2.0 g
Trypticase.....	2.0 g
Resazurin.....	1.0 mg
Cysteine HCl x H ₂ O.....	0.5 g
Na ₂ S x 9H ₂ O.....	0.5 g
DI Water to.....	1000 ml

This medium has problems with precipitate. Add chemicals in the order as listed. Make mg/ml solution for Fe(NH₄)₂(SO₄)₂ x 6H₂O, Resazurin, and Na₂S x 9H₂O. Gas under 80% N₂ and 20% CO₂ for 45 minutes. Check pH and lower to 7.0. Dispense, then autoclave.

Vitamin Solution

Biotin.....	2.0 mg
Folic Acid.....	2.0 mg
Pyridoxine x HCl.....	10.0 mg
Thiamine x HCl.....	5.0 mg
Riboflavin.....	5.0 mg
Nicotinic Acid.....	5.0 mg
Ca pantothenate.....	5.0 mg
Vitamin B ₁₂	0.01 mg
p-Aminobenzoic Acid.....	5.0 mg
Thioctic Acid.....	1.0 mg
DI Water.....	1000 ml

Filter Sterilize

Trace Element Solution SL-6

ZnSO ₄ x 7H ₂ O.....	0.1 g
MnCl ₂ x 4H ₂ O.....	0.03 g
H ₃ BO ₃	0.3 g
CoCl ₂ x 6H ₂ O.....	0.2 g

CuCl ₂ x 2H ₂ O.....	0.01 g
NiCl ₂ x 6H ₂ O.....	0.02 g
Na ₂ MoO ₄ x 2H ₂ O.....	0.03 g
DI Water.....	1000 ml

Filter sterilize