

ATCC medium: 2188 Modified VWN medium

Solution A:

KH₂PO₄0.2 g
NH₄Cl0.25 g
NaCl.....1.0 g
MgCl₂ . 6H₂O0.4 g
KCl.....0.5 g
CaCl₂ . 2H₂O0.15 g
Resazurin.....0.5 mg
Distilled water.....940.0 ml

SL-10 Trace Element Solution (see below).....1.0 ml
Wolfe's Vitamin Solution (see below).....10.0 ml
Selenite/Tungstate Solution (see below).....10.0 ml
Solution E (see below).....50.0 ml
Solution F (see below)
Solution G (see below)

Prepare Solution A anaerobically under 80% N₂, 20% CO₂. Autoclave at 121C for 15 minutes. Allow to cool under gas. Prepare and filter-sterilize anaerobic stock solutions of other solutions separately, then add them in order given to Solution A.

Adjust final pH to 7.2-7.4 if necessary.

SL-10 Trace Element Solution:

HCl (25%).....10.0 ml
FeCl₂ . 4H₂O1.5 g
ZnCl₂70.0 mg
MnCl₂ . 4H₂O100.0 mg
H₃BO₃6.0 mg
CoCl₂ . 6H₂O190.0 mg
CuCl₂ . 2H₂O2.0 mg
NiCl₂ . 6H₂O24.0 mg
Na₂MoO₄ . 2H₂O.....36.0 mg
Distilled water.....990.0 ml

Dissolve FeCl₂ in the HCl, dilute with water, add and dissolve the other salts; adjust pH to 6.0 with NaOH, and fill to 1.0 L with distilled water.

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

Selenite/Tungstate Solution:

NaOH.....0.5 g
Na₂SeO₃ . 5H₂O.....3.0 mg
Na₂WO₄ . 2H₂O.....4.0 mg
Distilled water.....100.0 ml

Solution E:

NaHCO₃ (5% w/v) solution in water

Solution F:

Use 2.5 g/L taurine as substrate (from anaerobic, 6.25% (v/v) stock solution)

Solution G:

Na₂S . 9H₂O0.3 g
Distilled water.....10.0 ml