ATCC medium: 1476 TYEG medium

NaCl..............................100.0 g
NH₄Cl..............................1.0 g
KH₂PO₄..............................0.3 g
Na₂HPO₄ . 7H₂O......................2.1 g
MgCl₂ . 6H₂O........................0.2 g
Trypticase Peptone (BD 211921)......10.0 g
Trace Mineral Solution II (see below)...10.0 ml
2.5% FeSO₄........................25.0 ml
Resazurin..........................2.0 mg
Distilled water to..............960.0 ml

Adjust medium to pH 7.3, +/- 0.1. Bring to boiling under 90% N₂, 10% CO₂
and autoclave under the same atmosphere at 121°C for 15 minutes.
Cool and aseptically add the following reduced filter-sterilized
solutions:

10% Glucose..........................50.0 ml
10% Yeast extract.......................5.0 ml
Wolfe's Vitamin Solution (see below)...10.0 ml

At time of inoculation add 0.125 ml of 2.5% Na₂S . 9H₂O per 5 ml medium
for a final concentration of approximately 0.06% (g/100 ml of medium).

Trace Mineral Solution II:
Nitrilotriacetic acid..............12.8 g
FeSO₄ . 7H₂O.........................0.1 g
MnCl₂ . 4H₂O........................0.1 g
CoCl₂ . 6H₂O........................0.17 g
CaCl₂ . 2H₂O........................0.1 g
ZnCl₂................................0.1 g
CuCl₂ . 2H₂O.........................0.02 g
H₃BO₃.................................0.01 g
Na₂MoO₄ . 2H₂O......................0.01 g
NaCl..................................0.1 g
Na₂SeO₃.............................0.017 g
NiSO₄ . 6H₂O.........................0.026 g
Distilled water to..............1.0 L

Add nitrilotriacetic acid to 200 ml distilled water and adjust pH to 6.5
with KOH. Add about 600 ml distilled water and dissolve the remaining
components, in order, one at a time. Bring volume to 1.0 L with
distilled water. Remove any brown precipitate by filtration. Store under
N₂.
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