

ATCC medium: 2223 Anaerobic serine/arginine medium

NH ₄ Cl	0.3 g
K ₂ HPO ₄	0.2 g
KH ₂ PO ₄	0.3 g
MgCl ₂ . 6H ₂ O	0.4 g
CaCl ₂ . 2H ₂ O	0.15 g
KCl.....	0.5 g
NaCl.....	1.0 g
SL-10 Trace Element Solution (see below).....	1.0 ml
Selenite/Tungstate Solution (see below).....	1.0 ml
Yeast extract.....	2.0 g
Serine*.....	1.05 g
Resazurin.....	0.5 mg
Distilled water.....	940.0 ml

* Arginine-HCl, 1.74 g, may be used in place of the serine
Prepare medium anaerobically under 80% N₂, 20% CO₂. Autoclave at 121C
for 15 minutes. Cool under the same gas mixture. After cooling, add the
following filter-sterilized ingredients:

Na₂S . 9H₂O Solution (see below)
NaHCO₃ Solution (see below)
Wolfe's Vitamin Solution (see below)10.0 ml

Adjust final pH of the completed medium to 7.2. Dispense anaerobically
as desired.

SL-10 Trace Element Solution:

HCl (25%).....	10.0 ml
FeCl ₂ . 4H ₂ O	1.5 g
ZnCl ₂	70.0 mg
MnCl ₂ . 4H ₂ O	100.0 mg
H ₃ BO ₃	6.0 mg
CoCl ₂ . 6H ₂ O	190.0 mg
CuCl ₂ . 2H ₂ O	2.0 mg
NiCl ₂ . 6H ₂ O	24.0 mg
Na ₂ MoO ₄ . 2H ₂ O.....	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.

Selenite/Tungstate Solution:

NaOH.....	0.5 g
Na ₂ SeO ₃ . 5H ₂ O.....	3.0 mg
Na ₂ WO ₄ . 2H ₂ O.....	4.0 mg
Distilled water.....	1.0 L

Na₂S . 9H₂O Solution:

Na ₂ S . 9H ₂ O	0.6 g
Distilled water.....	10.0 ml

NaHCO₃ Solution:

NaHCO ₃	5.0 g
Distilled water.....	50.0 ml

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