

ATCC medium: 1648 Modified *Desulfobacterium* medium

Solution A:

Na₂SO₄3.0 g
KH₂PO₄0.2 g
NH₄Cl0.3 g
NaCl.....21.0 g
MgCl₂ . 6H₂O3.0 g
KCl.....0.5 g
CaCl₂ . 2H₂O0.15 g
Resazurin.....1.0 mg
Distilled water.....920.0 ml

Solution B:

Trace Element Solution SL-10 (see below).....1.0 ml

Solution C:

NaHCO₃2.5 g
Distilled water.....50.0 ml

Solution D:

Sodium acetate . 3H₂O2.5 g
Distilled water.....10.0 ml

Solution E:

Wolfe's Vitamin solution (see below).....10.0 ml

Solution F:

Na₂SeO . 5H₂O3.0 mg
0.01 M NaOH.....1.0 L

Use 1.0 ml/L medium.

Solution G:

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

Directions for complete medium:

Prepare and autoclave Solutions A and D anaerobically under 80% N₂, 20% CO₂. Filter-sterilize and gas Solution C for 20 minutes with 80% N₂, 20% CO₂. Filter-sterilize and gas Solutions B, E and F with N₂. Autoclave Solution G under N₂. Add Solutions B through G to the sterile, cooled Solution A in the sequence indicated. Final pH of the medium should be 7.0.

Addition of 10-20 mg/L sodium dithionite (e.g., from 5% (w/v) solution freshly prepared under N₂ and filter-sterilized) may stimulate growth of all strains at the beginning. For transfers use 5-10% inoculum. Incubate all strains in the dark.

Trace Elements Solution SL-10:

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.

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