

ATCC Medium: 2848 Geoglobus Medium

Nutrient Solution

NaCl.....	18.00 g
MgCl ₂ x 6H ₂ O.....	4.00 g
KCl.....	0.33 g
CaCl ₂ x 2H ₂ O.....	0.33 g
(NH ₄) ₂ SO ₄	0.50 g
KH ₂ PO ₄	0.33 g
Trace Element Solution SL-10 (see below).....	1.00 ml
Selenite-tungstate solution (see below).....	1.00 ml
Yeast Extract.....	0.20 g
Vitamin solution (see medium 141).....	1.00 ml
NaHCO ₃	2.50 g
DI Water.....	1000.00 ml

Dissolve ingredients except bicarbonate, hydrogen phosphate, yeast extract, and vitamins. Prepare the nutrient solution anoxically under 80% N₂ + 20% CO₂ gas mixture, mix 400 ml of nutrient solution with solid Fe(III)oxide, dispense in anoxic vials and autoclave at 121C. Add vitamin solution, hydrogenphosphate and yeast extract from sterile, anoxic stock solutions prepared under N₂ and bicarbonate from a sterile, anoxic stock solution prepared under 80% N₂ and 20% CO₂ gas mixture. The pH of the complete medium should be at **6.5 – 6.8**. Prepare medium under 80% H₂ and 20% CO₂ gas atmosphere and supplement medium upon autoclaving with 3.5 g/l Na₂S₂O₃ x 5 H₂O added from a sterile anoxic stock solution sterilized by filtration.

Amorphous Fe(III)oxide (solid)

Slowly titrate 160 ml of a FeCl₃ x 6 H₂O stock solution (60 g/l) with 10% (w/v) NaOH to pH 8.0-8.5 under agitation (use magnetic stirrer). Total amount of added NaOH ca. 40 – 50 ml. The precipitated Fe(OH)₃ should be stored at room temperature overnight with surface covered with water. Thereafter, centrifuge at 2000 rpm for 5 min. and discard the supernatant. Resuspend the pellet in nutrient solution as described below.

Trace element solution SL-10

HCl (25%; 7.7 M).....	10.00 ml
FeCl ₂ x 4H ₂ O.....	1.50 g
ZnCl ₂	70.00 mg
MnCl ₂ x 4H ₂ O.....	100.00 mg
H ₃ BO ₃	6.00 mg
CoCl ₂ x 6H ₂ O.....	190.00 mg
CuCl ₂ x 2H ₂ O.....	2.00 mg
NiCl ₂ x 6H ₂ O.....	24.00 mg
Na ₂ MoO ₄ x 2H ₂ O.....	36.00 mg
DI Water.....	990.00 ml

First dissolve FeCl₂ in the HCl, then dilute in water, add and dissolve the other salts.

Finally make up to 1000.0 ml.

Selenite-tungstate Solution

NaOH.....	0.5 g
Na ₂ SeO ₃ x 5 H ₂ O.....	3 mg
Na ₂ WO ₄ x 2 H ₂ O.....	4 mg
DI Water.....	1000 ml

Extraneous Substrates to be added:

0.5 ml/10 ml of 260 mM FeCl₂