

81: MINERAL MEDIUM FOR CHEMOLITHOTROPHIC GROWTH (H-3)

Final pH: 6.8

Final volume: 995 ml

Solution A	50.00	ml
Solution B	920.00	ml
Solution C	20.00	ml
Standard vitamin solution	5.00	ml

1. Solutions A, B, C are autoclaved separately for 15 min at 121°C, cooled down to 50°C and then mixed aseptically with 5.0 ml filter-sterilized standard vitamin solution (see below). The final pH of this medium should be 6.8 without adjustment.
2. For chemolithotrophic growth incubate the culture under an atmosphere of 2% (v/v) O₂, 10% CO₂, 60% H₂ and 28% N₂. For heterotrophic growth supplement the mineral medium with an appropriate carbon source (0.2% carbohydrate or 0.1% organic acid). For growth on nitrogen-free medium, omit NH₄Cl and incubate the culture under an atmosphere of 2% (v/v) O₂, 10% CO₂, 10% H₂ and 78% N₂ or heterotrophically under 2% (v/v) O₂ and 98% N₂. For more details see Ref. 1515 and Ref. 3363.

Solution A

KH ₂ PO ₄	2.30	g
Na ₂ HPO ₄ x 2 H ₂ O	2.90	g
Distilled water	50.00	ml

Solution B

NH ₄ Cl	1.00	g
MgSO ₄ x 7 H ₂ O	0.50	g
CaCl ₂ x 2 H ₂ O	0.01	g
MnCl ₂ x 4 H ₂ O	5.00	mg
NaVO ₃ x H ₂ O	5.00	mg
Trace element solution SL-6	5.00	ml
Distilled water	915.00	ml
Agar, if required	20.00	g

Solution C

Ferric ammonium citrate	0.05	g
Distilled water	20.00	ml

Standard vitamin solution (from medium 81)

Riboflavin	10.00	mg
Thiamine-HCl x 2 H ₂ O	50.00	mg
Nicotinic acid	50.00	mg
Pyridoxine hydrochloride	50.00	mg
Calcium pantothenate	50.00	mg
Biotin	0.10	mg
Folic acid	0.20	mg
Vitamin B ₁₂	1.00	mg
Distilled water	100.00	ml

Trace element solution SL-6 (from medium 27)

ZnSO ₄ x 7 H ₂ O	0.10	g
MnCl ₂ x 4 H ₂ O	0.03	g
H ₃ BO ₃	0.30	g
CoCl ₂ x 6 H ₂ O	0.20	g
CuCl ₂ x 2 H ₂ O	0.01	g
NiCl ₂ x 6 H ₂ O	0.02	g
Na ₂ MoO ₄ x 2 H ₂ O	0.03	g
Distilled water	1000.00	ml