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

Solution A:

KH_2PO_4	2.300	g
Na_2HPO_4 x 2 H_2O	2.900	g
Distilled water	50.000	ml

Solution B:

NH_4Cl	1.000	g
$MgSO_4$ x 7 H_2O	0.500	g
$CaCl_2$ x 2 H_2O	0.010	g
$MnCl_2$ x 4 H_2O	0.005	g
$NaVO_3$ x H_2O	0.005	g
Trace element sol. SL-6 (see medium 27)	5.000	ml
Distilled water	915.000	ml
Agar (if necessary)	20.000	g

Solution C:

Ferric ammonium citrate	0.050	g
Distilled water	20.000	ml

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.

For chemolithotrophic growth incubate the culture under an atmosphere of 2% (v/v) O_2, 10% CO_2, 60% H_2 and 28% N_2. 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 NH_4Cl and incubate the culture under an atmosphere of 2% (v/v) O_2, 10% CO_2, 10% H_2 and 78% N_2 or heterotrophically under 2% (v/v) O_2 and 98% N_2. For more details see Ref. 1515 and Ref. 3363.

Standard vitamin solution:

Riboflavin	10.000	mg
Thiamine-HCl x 2 H_2O	50.000	mg
Nicotinic acid	50.000	mg
Pyridoxine-HCl	50.000	mg
Ca-pantothenate	50.000	mg
Biotin	0.100	mg
Folic acid	0.200	mg
Vitamin B_{12}	1.000	mg
Distilled water	100.000	ml