

1479. HALODESULFURIARCHAEUM MEDIUM

NaCl	240.00	g
K ₂ HPO ₄	2.50	g
(NH ₄) ₂ SO ₄	0.50	g
HEPES	10.00	g
Trace element solution (see below)	1.00	ml
sulfur powder	2.00	g
Distilled water	1000.00	ml

Prepare the liquid medium. Dispense the medium into Hungate tubes or serum bottles under gassing with N₂. Steam medium for 3h on each of 3 successive days. Before inoculation add from sterile stock solutions:

3.00 mM	MgCl ₂ ·6H ₂ O
5.00 mM	NaHCO ₃
50.0 mM	sodium formate
0.20 g/l	yeast extract
1.00 ml/l	Vitamin mix (pfennig & Lippert,1966) see below
1.00 mM	sodium sulfide from 1M filter sterilized stock

Trace element solution:

EDTA	5.00	g
FeSO ₄ x 7 H ₂ O	2.00	g
ZnSO ₄	0.10	g
MnCl ₂	0.03	g
H ₃ BO ₃	0.30	g
CoCl ₂ x 6 H ₂ O	0.20	g
CuCl ₂ x 5 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	0.02	g
Distilled water	1000.00	ml

Final pH 3.0-4.0 (with HCl).

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Vitamin mix (pfennig & Lippert, 1966)

1: acidic solution (HCl, pH3; 100ml)

Thiamin	5.0	mg
Ca Pantotenate	5.0	mg

2: basic solution (100ml 0.1N NaOH)

Biotin	2.0	mg
PABA (p-aminobenzoic acid)	5.0	mg
Nicotinic acid	5.0	mg
Pyridoxine	10.0	mg

3: Neutral solution in 100 ml demi water

Folic acid	2.0	mg
Riboflavin	5.0	mg

4: neutral solution in 100 ml demi water

B12	0.5	mg
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Sterilize by filtration.

Mix 1:1:1:1 just after addition or add separately, each 1ml/L.