

1058b: HALOTHIOBACILLUS HL2 MEDIUM

NaCl	120.00	g
K ₂ HPO ₄	1.50	g
NH ₄ Cl	0.50	g
Trace elements solution (Pfennig & Lippert,1966)	1.00	ml
CaCl ₂ x 2 H ₂ O	0.05	g
MgCl ₂ x 6 H ₂ O	0.40	g
Na ₂ S ₂ O ₃ x 5 H ₂ O	5.00	g
NaHCO ₃	2.50	g
Seven vitamins solution	1.00	ml
Distilled water	1000.00	ml

1. Dissolve sodium chloride, potassium hydrogenphosphate and ammonium chloride, then sparge solution with 80% N₂ and 20% CO₂ gas mixture for at least 30 - 45 min to remove dissolved oxygen and to saturate the solution with CO₂. Dispense solution under air atmosphere in Erlenmeyer flasks to 10% of volume, close with screw caps and autoclave. Add trace elements, calcium chloride, magnesium chloride, thiosulfate, bicarbonate and vitamins from sterile stock solutions and adjust pH of the medium to 7.5 using a sterile stock solution of sodium carbonate (5% w/v). Incubate with shaking.

2. Note: Use at least 10% (v/v) as inoculum.

Trace elements solution (Pfennig & Lippert,1966) (from medium 1369)

EDTA	5.00	g
FeSO ₄ x 7 H ₂ O	2.20	g
ZnSO ₄ x 7 H ₂ O	0.10	g
MnCl ₂ x 4 H ₂ O	0.03	g
H ₃ BO ₃	0.03	g
CoCl ₂ x 6 H ₂ O	0.20	g
CuCl ₂ x 2 H ₂ O	0.03	g
NiCl ₂ x 6 H ₂ O	0.03	g
Na ₂ MoO ₄ x 2 H ₂ O	0.03	g
Distilled water	1000.00	ml

pH 3.0-4.0

Seven vitamins solution (from medium 503)

Vitamin B ₁₂	100.00	mg
p-Aminobenzoic acid	80.00	mg
D-(+)-biotin	20.00	mg
Nicotinic acid	200.00	mg
Calcium pantothenate	100.00	mg



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Pyridoxine hydrochloride	300.00	mg
Thiamine-HCl x 2 H ₂ O	200.00	mg
Distilled water	1000.00	ml