

818: CHRYSIOGENES MEDIUM

This recipe contains strain-specific modifications for *Alkalihalobacillus macyae* DSM 16346 *

Final pH: 7.4 - 7.8

Final volume: 1003 ml

NaCl	1.20	g
KCl	0.30	g
NH ₄ Cl	0.30	g
KH ₂ PO ₄	0.20	g
Na ₂ SO ₄	0.30	g
MgCl ₂ x 6 H ₂ O	0.40	g
CaCl ₂ x 2 H ₂ O	0.15	g
Trace element solution SL-11	1.00	ml
Sodium resazurin (0.1% w/v)	0.50	ml
NaHCO ₃	0.60	g
KNO ₃	0.50	g
Wolin's vitamin solution (10x)	1.00	ml
Seven vitamins solution	1.00	ml
Na-acetate	0.50	g
Yeast extract	0.80	g
Na-(L)-lactate	1.10	g
Distilled water	1000.00	ml

Dissolve ingredients except bicarbonate, nitrate, vitamins and acetate. Sparge medium with 100% N₂ gas for 30 - 45 min to make it anoxic, adjust pH to 7.0 - 7.4, then dispense under same gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. Add nitrate, vitamins and acetate from sterile anoxic stock solutions prepared under 100% N₂ gas and bicarbonate from a sterile anoxic stock solution prepared under 80% N₂ and 20% CO₂ gas atmosphere. Vitamins should be sterilized by filtration. The pH of the complete medium should be adjusted to 7.4 - 7.8, if necessary.

* Supplement medium with 0.80 g/l yeast extract and replace Na-acetate with 1.10 g/l Na-L-lactate.

Trace element solution SL-11 (from medium 722)

Na ₂ -EDTA x 2 H ₂ O	5.20	g
FeCl ₂ x 4 H ₂ O	1.50	g
ZnCl ₂	70.00	mg
MnCl ₂ x 4 H ₂ O	100.00	mg
H ₃ BO ₃	6.00	mg
CoCl ₂ x 6 H ₂ O	190.00	mg
CuCl ₂ x 2 H ₂ O	2.00	mg

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NiCl ₂ x 6 H ₂ O	24.00	mg
Na ₂ MoO ₄ x 2 H ₂ O	36.00	mg
Distilled water	1000.00	ml

Dissolve EDTA in 800 ml distilled water, adjust pH to 7 using 2 N NaOH and add ferrous chloride. After ferrous chloride has dissolved add other compounds. Finally adjust pH to 6.0 and bring volume to 1000 ml.

Wolin's vitamin solution (10x) (from medium 120)

Biotin	20.00	mg
Folic acid	20.00	mg
Pyridoxine hydrochloride	100.00	mg
Thiamine HCl	50.00	mg
Riboflavin	50.00	mg
Nicotinic acid	50.00	mg
Calcium D-(+)-pantothenate	50.00	mg
Vitamin B ₁₂	1.00	mg
p-Aminobenzoic acid	50.00	mg
(DL)-alpha-Lipoic acid	50.00	mg
Distilled water	1000.00	ml

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