

647: DESULFUROMONAS THIOPHILA MEDIUM

KH ₂ PO ₄	1.00	g
NH ₄ Cl	0.50	g
MgSO ₄ x 7 H ₂ O	0.40	g
CaCl ₂ x 2 H ₂ O	0.10	g
Trace element solution SL-10	1.00	ml
Selenite-tungstate solution	1.00	ml
Sodium resazurin (0.1% w/v)	0.50	ml
Sulfur (powdered)	5.00	g
Na ₂ CO ₃	1.50	g
Na-pyruvate	0.60	g
Seven vitamins solution	1.00	ml
Na ₂ S x 9 H ₂ O	0.50	g
Distilled water	1000.00	ml

Dissolve ingredients (except sulfur, carbonate, pyruvate, vitamins and sulfide), then sparge medium with 80% N₂ and 20% CO₂ gas mixture for 30 - 45 min to make it anoxic. Dispense medium under same gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. Sulfur is sterilized by steaming for 3 hours on each of 3 successive days (see medium 35) and added aseptically to the sterile medium while retaining anoxic conditions. Complete the medium by adding pyruvate, vitamins and sulfide from sterile anoxic stock solutions prepared under 100% N₂ gas atmosphere and carbonate from a sterile anoxic stock solution prepared under 80% N₂ and 20% CO₂ gas atmosphere. Stock solutions of pyruvate and vitamins are sterilized by filtration. Adjust pH of the complete medium to 7.0 - 7.2, if necessary.

Trace element solution SL-10 (from medium 320)

HCl (25%)	10.00	ml
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
NiCl ₂ x 6 H ₂ O	24.00	mg
Na ₂ MoO ₄ x 2 H ₂ O	36.00	mg
Distilled water	990.00	ml

First dissolve FeCl₂ in the HCl, then dilute in water, add and dissolve the other salts. Finally make up to 1000.00 ml.

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Selenite-tungstate solution (from medium 385)

NaOH	0.50	g
Na ₂ SeO ₃ x 5 H ₂ O	3.00	mg
Na ₂ WO ₄ x 2 H ₂ O	4.00	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