

647. DESULFUROMONAS THIOPHILA MEDIUM

KH ₂ PO ₄	1.0	g
NH ₄ Cl	0.5	g
MgSO ₄ x 7 H ₂ O	0.4	g
CaCl ₂ x 2 H ₂ O	0.1	g
Trace element solution SL-10 (see medium 320)	1.0	ml
Selenite-tungstate solution (see medium 385)	1.0	ml
Na-resazurin solution (0.1% w/v)	0.5	ml
Sulfur, powdered	5.0	g
NaHCO ₃	2.5	g
Na-pyruvate	0.6	g
Vitamin solution (see medium 503)	1.0	ml
Na ₂ S x 9 H ₂ O	0.5	g
Distilled water	1000.0	ml

Dissolve ingredients (except sulfur, bicarbonate, 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 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 bicarbonate 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.