

899. THERMOANAEROBACTER SUBTERRANEUS MEDIUM

NH ₄ Cl	1.00	g
K ₂ HPO ₄	0.30	g
KH ₂ PO ₄	0.30	g
MgCl ₂ x 6 H ₂ O	1.00	g
CaCl ₂ x 2 H ₂ O	0.10	g
KCl	0.20	g
NaCl	0.60	g
Trace element solution (see medium 141)	10.00	ml
Yeast extract	2.00	g
Na-resazurin solution (0.1% w/v)	0.50	ml
Na ₂ CO ₃	1.00	g
D-Glucose	4.00	g
Na ₂ S ₂ O ₃ x 5 H ₂ O	2.50	g
L-Cysteine-HCl x H ₂ O	0.50	g
Na ₂ S x 9 H ₂ O	0.45	g
Distilled water	1000.00	ml

Dissolve ingredients (except carbonate, glucose, thiosulfate, cysteine 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. Add glucose, thiosulfate (sterilized by filtration), cysteine and sulfide from sterile anoxic stock solutions prepared under 100% N₂ gas and carbonate from a sterile anoxic stock solution prepared under 80% N₂ and 20% CO₂ gas mixture. The pH of the complete medium should be at 7.0.