Microorganisms



Main sol. 853b

NH ₄ Cl	1.00	g
K ₂ HPO ₄	0.30	g
KH ₂ PO ₄	0.30	g
$MgCl_2 \times 6 H_2O$	0.50	g
CaCl ₂ x 2 H ₂ O	0.10	g
KCI	0.10	g
NaCl	3.00	g
Yeast extract (OXOID)	1.00	g
Trypticase peptone (BD BBL)	1.00	g
Modified Wolin's mineral solution	10.00	ml
Sodium resazurin (0.1% w/v)	0.50	ml
$Na_2S_2O_3 \times 5 H_2O$	3.16	g
L-Cysteine HCl x H ₂ O	0.50	g
Na ₂ CO ₃	1.50	g
D-Glucose	3.60	g
$Na_2S \times 9 H_2O$	0.30	g
Distilled water	1000.00	ml

Dissolve ingredients, except thiosulfate, cysteine, carbonate, glucose, and sulfide, then sparge medium for 30 - 45 min with 80% N_2 and 20% CO_2 gas mixture to make it anoxic. Add thiosulfate and cysteine, then adjust pH to 7.0 and dispense medium under 80% N_2 and 20% CO_2 gas atmosphere into anoxic Hungate-type tubes or serum vials to 30% of their volume and autoclave. After autoclaving, add glucose and sulfide from sterile anoxic stock solutions prepared under 100% N_2 gas and carbonate from a sterile anoxic stock solution prepared under 80% N_2 and 20% CO_2 gas atmosphere. Adjust the pH of the complete medium to 7.2 - 7.4, if necessary.