

Main sol. 326

KH ₂ PO ₄	3.30	g
MgCl ₂ x 6 H ₂ O	0.16	g
CoCl ₂ x 6 H ₂ O	12.00	mg
Trace element solution SL-10	1.00	ml
Yeast extract	2.00	g
Trypticase peptone (BD BBL)	2.00	g
Sodium resazurin (0.1% w/v)	0.50	ml
Na ₂ CO ₃	1.00	g
Sucrose	5.00	g
Wolin's vitamin solution (10x)	1.00	ml
L-Cysteine HCl x H ₂ O	0.30	g
Na ₂ S x 9 H ₂ O	0.30	g
Distilled water	1000.00	ml

Dissolve ingredients (except carbonate, sucrose, vitamins, cysteine and sulfide) and 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. After autoclaving add sucrose, cysteine and sulfide to the medium from sterile stock solutions autoclaved under 100% N₂ gas and carbonate from a sterile anoxic stock solution prepared under 80% N₂ and 20% CO₂ gas mixture. Vitamins are prepared under 100% N₂ gas and sterilized by filtration. Adjust pH of complete medium to pH 6.8 - 7.2, if necessary.