

Main sol. 1206

KH ₂ PO ₄	0.27	g
NH ₄ Cl	0.50	g
(NH ₄) ₂ SO ₄	0.50	g
TAPS sodium salt (SIGMA)	2.40	g
MgCl ₂ x 6 H ₂ O	0.01	g
CaCl ₂ x 2 H ₂ O	0.02	g
Yeast extract	1.00	g
Modified Wolin's mineral solution	10.00	ml
Sodium resazurin (0.1% w/v)	0.50	ml
NaHCO ₃	1.50	g
D-Xylose	3.00	g
Wolin's vitamin solution (10x)	1.00	ml
L-Cysteine HCl x H ₂ O	0.25	g
Na ₂ S x 9 H ₂ O	0.25	g
Distilled water	1000.00	ml

Dissolve ingredients (except bicarbonate, xylose, vitamins, cysteine and sulfide), adjust pH to 8.5, then sparge medium with 100% N₂ gas 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 xylose, vitamins, cysteine and sulfide from sterile anoxic stock solutions prepared under 100% N₂ gas and bicarbonate from a sterile anoxic stock solution prepared under 80% N₂ and 20% CO₂ gas atmosphere. Sterilize stock solutions of xylose and vitamins by filtration. Adjust pH of the complete medium to 8.5, if necessary.