Main sol. 318b

KH_2PO_4	0.30	g
NaCl	0.60	g
$MgCl_2$ x 6 H_2O	0.10	g
$CaCl_2$ x 2 H_2O	0.08	g
Trace element solution	10.00	ml
NH_4Cl	1.00	g
Yeast extract (OXOID)	0.50	g
Trypticase peptone (BD BBL)	0.50	g
Sodium resazurin (0.1% w/v)	0.50	ml
$KHCO_3$	4.00	g
Ethanol	1.30	ml
Wolin's vitamin solution (10x)	1.00	ml
L-Cysteine HCl x H_2O	0.30	g
Na_2S x 9 H_2O	0.30	g
Distilled water	1000.00	ml

Dissolve ingredients (except bicarbonate, ethanol, vitamins, cysteine and sulfide), then sparge medium with 80% N_2 and 20% CO_2 gas mixture for 30 - 45 min to make it anoxic. Add and dissolve bicarbonate and adjust pH to 7.0, then dispense under 80% N_2 and 20% CO_2 gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. After sterilization add ethanol, cysteine and sulfide from sterile anoxic stock solutions prepared under 100% N_2 gas. Vitamins are prepared under 100% N_2 gas and sterilized by filtration. Adjust pH of complete medium to 7.0 - 7.2, if necessary.