

Main sol. 503a

KH ₂ PO ₄	0.20	g
NH ₄ Cl	0.25	g
NaCl	1.00	g
MgCl ₂ x 6 H ₂ O	0.40	g
KCl	0.50	g
CaCl ₂ x 2 H ₂ O	0.15	g
Yeast extract (OXOID)	1.00	g
Casamino acids (BD Bacto)	1.00	g
Trace element solution SL-10	1.00	ml
Selenite-tungstate solution	1.00	ml
Sodium resazurin (0.1% w/v)	0.50	ml
Na ₂ CO ₃	1.50	g
Seven vitamins solution	1.00	ml
Na-DL-lactate	5.00	g
DL-Dithiothreitol (DTT)	3.00	g
Distilled water	1000.00	ml

Dissolve ingredients (except carbonate, vitamins, lactate and DTT), 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 complete the medium by adding vitamins, lactate and DTT from anoxic stock solutions prepared under 100% N₂ gas and carbonate from a sterile anoxic stock solution prepared under 80% N₂ and 20% CO₂ gas atmosphere. Vitamins are sterilized by filtration. Adjust pH of the complete medium to 7.0 - 7.2, if necessary.