

Main sol. 124

NaCl	1.17	g
MgCl ₂ x 6 H ₂ O	0.40	g
KCl	0.30	g
CaCl ₂ x 2 H ₂ O	0.15	g
NH ₄ Cl	0.27	g
KH ₂ PO ₄	0.20	g
Na ₂ SO ₄	2.84	g
Trace element solution SL-10	1.00	ml
Selenite-tungstate solution	1.00	ml
Yeast extract	1.00	g
Sodium resazurin (0.1% w/v)	0.50	ml
NaHCO ₃	4.50	g
Na-acetate	1.40	g
Na-butyrate	1.40	g
Wolin's vitamin solution (10x)	1.00	ml
Na ₂ S x 9 H ₂ O	0.40	g
Distilled water	1000.00	ml

Dissolve ingredients (except bicarbonate, acetate, butyrate, vitamins, and sulfide) and sparge medium with 80% N₂ and 20% CO₂ gas mixture for 30 - 45 min to make it anoxic. Add and dissolve bicarbonate, adjust pH to 7.0, then dispense medium under 80% N₂ and 20% CO₂ gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. Add acetate, butyrate, vitamins, and sulfide from sterile anoxic stock solutions prepared under 100% N₂ gas. The vitamin solution should be sterilized by filtration. Before use, check the complete medium's pH and adjust it to 7.0 - 7.2, if necessary.