

Main sol. J696

NaCl	20.00	g
MgCl ₂ x 6 H ₂ O	3.00	g
Na ₂ SO ₄	3.00	g
KCl	0.50	g
NH ₄ Cl	0.30	g
KH ₂ PO ₄	0.10	g
PIPES	1.73	g
CaCl ₂ x 2 H ₂ O	0.11	g
Trace minerals	10.00	ml
Trace vitamins	10.00	ml
Na ₂ S x 9 H ₂ O	0.30	g
Distilled water	1000.00	ml

Mix components except Na₂S x 9H₂O and adjust pH to 6.7. Distribute the medium into culture vessels (e.g., 20 ml in 120 ml serum bottles) under a gas stream of H₂-CO₂ (4:1, v/v), seal with butyl rubber stoppers and autoclave. Separately autoclave 3% Na₂S x 9H₂O solution under a N₂ gas atmosphere. Prior to inoculation, reduce the medium by the addition of the Na₂S x 9H₂O solution. Pressurized the inoculated culture vessels to 200 kPa H₂-CO₂ (4:1, v/v).