

135a. ACETOBACTERIUM MEDIUM (MARINE)

NaCl	20.00	g
NH ₄ Cl	1.00	g
KH ₂ PO ₄	0.33	g
K ₂ HPO ₄	0.45	g
MgSO ₄ x 7 H ₂ O	0.10	g
Trace element solution (see medium 141)	20.00	ml
Yeast extract	2.00	g
Resazurin solution (0.1% w/v)	0.50	ml
NaHCO ₃	5.00	g
Ethylene glycol	1.25	g
Vitamin solution (see medium 141)	10.00	ml
L-Cysteine-HCl x H ₂ O	0.50	g
Na ₂ S x 9 H ₂ O	0.50	g
Distilled water	1000.00	ml

Dissolve ingredients except bicarbonate, ethylene glycol, vitamins, cysteine and sulfide, bring to the boil and cool to room temperature under 80% N₂ and 20% CO₂ gas mixture. Add bicarbonate (solid) and equilibrate the medium with the gas until a pH of around 7.4 is reached. Then distribute under the same gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. Before use add ethylene glycol, vitamins (sterilized by filtration), cysteine and sulfide from sterile anoxic stock solutions prepared under 100% N₂ gas. Adjust pH of complete medium to 7.0 – 7.2, if necessary.