

1329. METHANOSAETA PELAGICA MEDIUM

MgCl ₂ x 6 H ₂ O	3.05	g
CaCl ₂ x 2 H ₂ O	0.15	g
NH ₄ Cl	0.54	g
NaCl	20.00	g
Na-acetate	6.56	g
Trace elements solution (see medium 318)	2.00	ml
Yeast extract (OXOID)	1.50	g
Trypticase peptone (BD BBL)	0.40	g
Na-resazurin solution (0.1% w/v)	0.50	ml
KH ₂ PO ₄	1.19	g
K ₂ HPO ₄	0.21	g
Na ₂ CO ₃	1.50	g
2-Mercaptoethanesulfonic acid (coenzyme M)	0.14	g
Vitamins solution (see medium 141)	10.00	ml
Na ₂ S x 9 H ₂ O	0.36	g
Distilled water	1000.00	ml

Dissolve ingredients (except phosphates, carbonate, coenzyme M, vitamins and sulfide) and 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. Add phosphates, vitamins (sterilized by filtration) and sulfide from sterile anoxic stock solutions prepared under 100% N₂ gas and carbonate from a sterile anoxic stock solution prepared under 80% N₂ and 20% CO₂ gas mixture. Adjust pH of the complete medium to 7.5, if necessary.