

348. METHANOHALOPHILUS PORTUCALENSIS MEDIUM

NaCl	120.00	g
MgCl ₂ x 6 H ₂ O	7.00	g
MgSO ₄ x 7 H ₂ O	6.00	g
CaCl ₂ x 2 H ₂ O	0.50	g
K ₂ HPO ₄ x 3 H ₂ O	0.40	g
NH ₄ Cl	1.00	g
KCl	3.80	g
Trace element solution SL-10 (see medium 320)	1.00	ml
Selenite-tungstate solution (see medium 385)	1.00	ml
Yeast extract (OXOID)	2.00	g
Trypticase peptone (BD BBL)	2.00	g
Na-resazurin solution (0.1% w/v)	0.50	ml
L-Cysteine-HCl x H ₂ O	0.50	g
Na ₂ CO ₃	1.50	g
Trimethylamine-HCl	1.90	g
Na ₂ S x 9 H ₂ O	0.25	g
Distilled water	1000.00	ml

Dissolve ingredients except cysteine, carbonate, trimethylamine and sulfide, bring medium to the boil, then cool to room temperature while sparging with 80% N₂ and 20% CO₂ gas mixture. Add cysteine and adjust pH to 6.5, then dispense under 80% N₂ and 20% CO₂ gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. Thereafter, add trimethylamine 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 complete medium to 7.3 - 7.4, if necessary.