Microorganisms



Main sol. 720

NH ₄ Cl	1.00	g
NaCl	0.10	g
$MgCl_2 \times 6 H_2O$	0.10	g
CaCl ₂ x 2 H ₂ O	0.05	g
$K_2HPO_4 \times 3 H_2O$	0.40	g
Trace element solution SL-11	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	2.60	g
Na-pyruvate	2.50	g
$Na_2S_2O_3 \times 5 H_2O$	1.25	g
Wolin's vitamin solution (10x)	1.00	ml
$Na_2S \times 9 H_2O$	0.30	g
Distilled water	1000.00	ml

Dissolve ingredients (except bicarbonate, pyruvate, thiosulfate, vitamins and sulfide), then sparge medium with $80\%~N_2$ and $20\%~CO_2$ gas mixture for 30 - 45 min to make it anoxic. Add bicarbonate and adjust pH to 7.0 - 7.2, then dispense under $80\%~N_2$ and $20\%~CO_2$ gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. Prior to use add pyruvate, thiosulfate, vitamins, and sulfide from sterile anoxic stock solutions prepared under $100\%~N_2$ gas. Stock solutions of pyruvate and vitamins are sterilized by filtration.