Microorganisms



870. SYNTROPHOTHERMUS MEDIUM

KH ₂ PO ₄	0.14	g
MgCl ₂ x 6 H ₂ O	0.20	g
CaCl ₂ x 2 H ₂ O	0.15	g
NH_4CI	0.54	g
Trace element solution (see medium 141)	1.00	ml
Na-resazurin solution (0.1% w/v)	0.50	ml
Na_2CO_3	1.00	g
Na-crotonate solution (1 M, see below)	10.00	ml
Vitamin solution (see medium 141)	10.00	ml
Na ₂ S x 9 H ₂ O	0.30	g
L-Cysteine-HCl x H₂O	0.30	g
Distilled water	1000.00	ml

Dissolve all ingredients except carbonate, crotonate, vitamins, sulfide and cysteine, then sparge medium with $80\%~N_2$ and $20\%~CO_2$ 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 crotonate, vitamins, sulfide and cysteine from sterile anoxic stock solutions prepared under $100\%~N_2$ gas and carbonate from a sterile anoxic stock solution prepared under $80\%~N_2$ and $20\%~CO_2$ gas atmosphere. Sterilize vitamins by filtration. Adjust pH of complete medium to 7.0, if necessary.

Na-crotonate solution (1 M):

Crotonic acid (ALDRICH 113018)	0.86	g
10 N NaOH	1.00	ml
Distilled water	9.00	ml

Dissolve crotonic acid in 8 ml distilled water, add around 1 ml of 10 N NaOH and adjust pH to around 7. Then add water to reach a volume of 10 ml. Sterilize by filtration under $100\% N_2$ gas atmosphere.

For <u>DSM 12681</u> replace Na-crotonate with 2.20 g/l Na-butyrate.