880: THERMOACETOGENIUM MEDIUM

KH_2PO_4	0.30	g
NaCl	0.60	g
$MgCl_2$ x 6 H_2O	0.10	g
$CaCl_2$ x 2 H_2O	0.08	g
NH_4Cl	1.00	g
Trace element solution SL-11	1.00	ml
Selenite-tungstate solution	1.00	ml
Sodium resazurin (0.1% w/v)	0.50	ml
$KHCO_3$	3.50	g
L-Cysteine HCl x H_2O	0.60	g
Methanol (50% v/v)	6.00	ml
Wolin's vitamin solution (10x)	1.00	ml
Na_2S x 9 H_2O	0.30	g
Distilled water	1000.00	ml

Dissolve ingredients (except bicarbonate, cysteine, methanol, vitamins and sulfide), sparge medium with 80% N_2 and 20% CO_2 gas mixture for 30 - 45 min to make it anoxic, then add solid bicarbonate and cysteine and equilibrate the medium with the gas mixture to reach a pH of 7.0. Dispense medium under 80% N_2 and 20% CO_2 gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. To the autoclaved medium add methanol (50% v/v solution), vitamins and sulfide from sterile anoxic stock solutions prepared under 100% N_2 gas. Vitamins should be sterilized by filtration. Adjust pH of complete medium to 7.0 - 7.2, if necessary.

Trace element solution SL-11 (from medium 722)

Na_2-EDTA x 2 H_2O	5.20	g
$FeCl_2$ x 4 H_2O	1.50	g
$ZnCl_2$	70.00	mg
$MnCl_2$ x 4 H_2O	100.00	mg
H_3BO_3	6.00	mg
$CoCl_2$ x 6 H_2O	190.00	mg
$CuCl_2$ x 2 H_2O	2.00	mg
$NiCl_2$ x 6 H_2O	24.00	mg
Na_2MoO_4 x 2 H_2O	36.00	mg
Distilled water	1000.00	ml

Dissolve EDTA in 800 ml distilled water, adjust pH to 7 using 2 N NaOH and add ferrous chloride. After ferrous chloride has dissolved add other compounds. Finally adjust pH to 6.0 and bring volume to 1000 ml.

Selenite-tungstate solution (from medium 385)

NaOH	0.50	g
Na_2SeO_3 x 5 H_2O	3.00	mg
Na_2WO_4 x 2 H_2O	4.00	mg
Distilled water	1000.00	ml

Wolin's vitamin solution (10x) (from medium 120)

Biotin	20.00	mg
Folic acid	20.00	mg
Pyridoxine hydrochloride	100.00	mg
Thiamine HCl	50.00	mg
Riboflavin	50.00	mg
Nicotinic acid	50.00	mg
Calcium D-(+)-pantothenate	50.00	mg
Vitamin B_{12}	1.00	mg
p-Aminobenzoic acid	50.00	mg
(DL)-alpha-Lipoic acid	50.00	mg
Distilled water	1000.00	ml