1361. PLANCTOMYCETALES BACTERIUM MEDIUM

Mineral Salts Solution:

KH_2PO_4	0.14	g
$MgCl_2$ x $6H_20$	0.20	g
$CaCl_2$ x 2 H_2O	0.15	g
NH_4Cl	0.54	g
Trace Elements (see below)	1.00	ml
Glucose	0.18	g
Yeast Extract	0.20	g
Vitamin Solution (see medium141) (see below)	1.00	ml
Resazurin (0.1 v/v)	0.50	ml
$NaHCO_3$	2.50	g
Na_2S 9 H_2O	0.30	g
Cysteine HCl	0.30	g
Distilled Water	1000.00	ml

Vitamin Solution 141:

Biotin	2.00	mg
Folic acid	2.00	mg
Pyridoxine-HCl	10.00	mg
Thiamine-HCl x 2 H_2O	5.00	mg
Riboflavin	5.00	mg
Nicotinic acid	5.00	mg
D-Ca-pantothenate	5.00	mg
Vitamin B_{12}	0.10	mg
p-Aminobenzoic acid	5.00	mg
Lipoic acid	5.00	mg
Distilled water	1000.00	ml

Trace elements Solution:

Nitrilotriacetic acid (NTA)	12.800	g
$FeCl_3$ x 6 H_2O	1.350	g
$MnCl_2$ x 4 H_2O	0.100	g
$CoCl_2$ x 6 H_2O	0.024	g
$CaCl_2$ x 2 H_2O	0.100	g
$ZnCl_2$	0.100	g
$CuCl_2$ x 2 H_2O	0.025	g
H_3BO_3	0.010	g
Na_2MoO_4 x 2 H_2O	0.024	g
$NiCl_2$ x 6 H_2O	0.120	g
Na_2SeO_3 x 5 H_2O	0.004	g
Distilled water	1000.000	ml

Prepare the medium without yeast extract, glucose, $NaHCO_3$, cysteine hydrochloride, vitamin solution and Na_2S x 9 H_2O. Boil and then cool under a gas atmosphere of N_2:CO_2 (80:20 v/v). Dispense the medium into Hungate tubes or serum bottles under gassing with N_2:CO_2 (80:20 v/v) and add the $NaHCO_3$. The final pH should be 7.0. Autoclave the medium and add from sterile stock solutions yeast extract, glucose, cysteine hydrochloride, vitamin solution and Na_2S x 9 H_2O.The final pH should be 7.0.