742: DESULFONATRONOVIBRIO MEDIUM

NaCl	10.00	g
Na_2SO_4	3.00	g
Na_2HPO_4	0.20	g
KCl	0.20	g
NH_4Cl	1.00	g
Modified Wolin's mineral solution	10.00	ml
Sodium resazurin (0.1% w/v)	0.50	ml
$NaHCO_3$	15.00	g
Na_2CO_3	10.00	g
Yeast extract	1.50	g
Na-formate	5.00	g
Wolin's vitamin solution (10x)	1.00	ml
Na_2S x 9 H_2O	1.00	g
Distilled water	1000.00	ml

Dissolve ingredients except bicarbonate, carbonate, yeast extract, vitamins, formate, and sulfide. Sparge medium with 100% N_2 gas for 30 - 45 min to make it anoxic, then add bicarbonate and carbonate and adjust pH to 9.5. Dispense medium under 100% N_2 gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. Add yeast extract, vitamins, formate and sulfide from sterile anoxic stock solutions prepared under 100% N_2 gas. Vitamins should be sterilized by filtration. The pH of the complete medium should be at 9.5 - 9.7.

Modified Wolin's mineral solution (from medium 141)

Nitrilotriacetic acid	1.50	g
$MgSO_4$ x 7 H_2O	3.00	g
$MnSO_4$ x H_2O	0.50	g
NaCl	1.00	g
$FeSO_4$ x 7 H_2O	0.10	g
$CoSO_4$ x 7 H_2O	0.18	g
$CaCl_2$ x 2 H_2O	0.10	g
$ZnSO_4$ x 7 H_2O	0.18	g
$CuSO_4$ x 5 H_2O	0.01	g
$AlK(SO_4)_2$ x 12 H_2O	0.02	g
H_3BO_3	0.01	g
Na_2MoO_4 x 2 H_2O	0.01	g
$NiCl_2$ x 6 H_2O	0.03	g
Na_2SeO_3 x 5 H_2O	0.30	mg
Na_2WO_4 x 2 H_2O	0.40	mg
Distilled water	1000.00	ml

First dissolve nitrilotriacetic acid and adjust pH to 6.5 with KOH, then add minerals. Adjust final to pH 7.0 with KOH.

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