819. SEDIMINISPIROCHAETA SMARAGDINAE MEDIUM

NH_4Cl	1.0	g
K_2HPO_4	0.3	g
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$MgCl_2$ x 6 H_2O	0.2	g
$CaCl_2$ x 2 H_2O	0.1	g
KCl	0.2	g
NaCl	50.0	g
Yeast extract	5.0	g
Trace element solution (see medium 141)	10.0	ml
Na-resazurin solution (0.1% w/v)	0.5	ml
L-Cysteine-HCl x H_2O	0.3	g
$NaHCO_3$	2.5	g
Na_2S x 9 H_2O	0.3	g
Distilled water	1000.0	ml

Dissolve ingredients (except cysteine, bicarbonate and sulfide), sparge medium with 80% N_2 and 20% CO_2 gas mixture for 30 – 45 min to make it anoxic. Then, add cysteine, distribute medium under 80% N_2 and 20% CO_2 gas atmosphere in anoxic Hungate-type tubes or serum vials and autoclave. After sterilization add sulfide from a sterile anoxic stock solution prepared under 100% N_2 gas atmosphere and bicarbonate from a sterile anoxic stock solution prepared under 80% N_2 and 20% CO_2 gas atmosphere. Check pH of complete medium and adjust to pH 7.0, if necessary.

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