

1263: SPIROCHAETA DISSIPATITROPHA MEDIUM

NaCl	30.00	g
KCl	0.20	g
K ₂ HPO ₄	0.20	g
MgCl ₂ × 6 H ₂ O	0.10	g
NH ₄ Cl	1.00	g
Modified Wolin's mineral solution	10.00	ml
Sodium resazurin (0.1% w/v)	0.50	ml
Na ₂ CO ₃	2.76	g
NaHCO ₃	24.00	g
Yeast extract	0.50	g
Cellobiose	5.00	g
Wolin's vitamin solution	2.00	ml
Na ₂ S × 9 H ₂ O	0.40	g
Distilled water	1000.00	ml

Dissolve ingredients (except carbonate, bicarbonate, yeast extract, cellobiose, vitamins and sulfide), then sparge medium with 100% N₂ gas for 30 - 45 min to make it anoxic. Add solid carbonate and bicarbonate to the medium, dissolve and adjust to pH 9.4. Dispense medium under 100% N₂ gas atmosphere into anoxic Hungate-type tubes or serum bottles and autoclave. To the autoclaved medium add yeast extract, cellobiose, vitamins and sulfide from sterile anoxic stock solutions prepared under 100% N₂ gas atmosphere. Stock solutions of vitamins and cellobiose should be sterilized by filtration.

For DSM 8901: For DSM 8901 exchange 'Modified Wolin's Mineral Solution' with 'Modified Wolin's Mineral Solution II'

Modified Wolin's mineral solution (from medium 141)

Nitrilotriacetic acid	1.50	g
MgSO ₄ × 7 H ₂ O	3.00	g
MnSO ₄ × H ₂ O	0.50	g
NaCl	1.00	g
FeSO ₄ × 7 H ₂ O	0.10	g
CoSO ₄ × 7 H ₂ O	0.18	g
CaCl ₂ × 2 H ₂ O	0.10	g
ZnSO ₄ × 7 H ₂ O	0.18	g
CuSO ₄ × 5 H ₂ O	0.01	g
AlK(SO ₄) ₂ × 12 H ₂ O	0.02	g
H ₃ BO ₃	0.01	g
Na ₂ MoO ₄ × 2 H ₂ O	0.01	g
NiCl ₂ × 6 H ₂ O	0.03	g
Na ₂ SeO ₃ × 5 H ₂ O	0.30	mg
Na ₂ WO ₄ × 2 H ₂ O	0.40	mg

Distilled water	1000.00	ml
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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 (from medium 141)

Biotin	2.00	mg
Folic acid	2.00	mg
Pyridoxine hydrochloride	10.00	mg
Thiamine HCl	5.00	mg
Riboflavin	5.00	mg
Nicotinic acid	5.00	mg
Calcium D-(+)-pantothenate	5.00	mg
Vitamin B ₁₂	0.10	mg
p-Aminobenzoic acid	5.00	mg
(DL)-alpha-Lipoic acid	5.00	mg
Distilled water	1000.00	ml