

282b: DESULFUROBACTERIUM HR11 MEDIUM

K ₂ HPO ₄	0.14	g
CaCl ₂ x 2 H ₂ O	0.14	g
NH ₄ Cl	0.25	g
MgSO ₄ x 7 H ₂ O	3.40	g
MgCl ₂ x 6 H ₂ O	4.10	g
KCl	0.33	g
NiCl ₂ x 6 H ₂ O (0.1% w/v)	0.50	ml
NaCl	30.00	g
Modified Wolin's mineral solution	10.00	ml
Sodium resazurin (0.1% w/v)	0.50	ml
NaHCO ₃	1.00	g
Na ₂ S ₂ O ₃ x 5 H ₂ O	1.00	g
Wolin's vitamin solution (10x)	1.00	ml
DL-Dithiothreitol (DTT)	1.00	g
Distilled water	1000.00	ml

1. Dissolve ingredients except bicarbonate, sodium thiosulfate, vitamins, and DTT. Sparge medium with 80% H₂ and 20% CO₂ gas mixture for 30 - 45 min to make it anoxic. Add and dissolve bicarbonate, then dispense medium into anoxic tubes to 30% of their volume under 80% H₂ and 20% CO₂ gas atmosphere and autoclave. Add thiosulfate, vitamins and DTT from anoxic stock solutions prepared under 100% N₂ gas and sterilized by filtration. Prior to use check pH of completed medium and adjust to 6.0, if necessary.
2. After inoculation add sterile 80% H₂ and 20% CO₂ gas mixture to 2 bar overpressure.

Modified Wolin's mineral solution (from medium 141)

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