

813b: DESULFONATRONUM LACUSTRE MEDIUM

This recipe contains strain-specific modifications for *Desulfobotulus pelophilus* DSM 112796 *

Final pH: 8.8 - 9.0

Final volume: 1003 ml

KH ₂ PO ₄	0.20	g
MgCl ₂ x 6 H ₂ O	0.10	g
NH ₄ Cl	1.00	g
KCl	0.20	g
Na ₂ SO ₄	5.00	g
NaCl	2.00	g
Trace element solution SL-10	1.00	ml
Selenite-tungstate solution	1.00	ml
Yeast extract	1.00	g
Sodium resazurin (0.1% w/v)	0.50	ml
Na ₂ CO ₃	3.50	g
Na ₂ S x 9 H ₂ O	0.50	g
Na-L-lactate	2.00	g
Wolin's vitamin solution (10x)	1.00	ml
Na-pyruvate	1.10	g
Distilled water	1000.00	ml

Dissolve ingredients (except carbonate, sulfide, lactate, and vitamins) and sparge medium with 100 % N₂ gas for 30 - 45 min to make it anoxic. Add and dissolve carbonate and sulfide, then adjust pH to 8.8 - 9.0, dispense under 100% N₂ gas atmosphere into Hungate-type tubes or serum vials and autoclave. Before use add lactate and vitamins (sterilized by filtration) from sterile anoxic stock solutions prepared under 100% N₂ gas.

* Supplement medium with 1.10 g Na-pyruvate added after autoclaving from a sterile anoxic stock solution sterilized by filtration. Omit Na-L-lactate.

Trace element solution SL-10 (from medium 320)

HCl (25%)	10.00	ml
FeCl ₂ x 4 H ₂ O	1.50	g
ZnCl ₂	70.00	mg
MnCl ₂ x 4 H ₂ O	100.00	mg
H ₃ BO ₃	6.00	mg
CoCl ₂ x 6 H ₂ O	190.00	mg
CuCl ₂ x 2 H ₂ O	2.00	mg
NiCl ₂ x 6 H ₂ O	24.00	mg
Na ₂ MoO ₄ x 2 H ₂ O	36.00	mg
Distilled water	990.00	ml

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First dissolve FeCl_2 in the HCl , then dilute in water, add and dissolve the other salts. Finally make up to 1000.00 ml.

Selenite-tungstate solution (from medium 385)

NaOH	0.50	g
$\text{Na}_2\text{SeO}_3 \times 5 \text{ H}_2\text{O}$	3.00	mg
$\text{Na}_2\text{WO}_4 \times 2 \text{ H}_2\text{O}$	4.00	mg
Distilled water	1000.00	ml

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