

193: DESULFOBACTER MEDIUM

This recipe contains strain-specific modifications for *Garciella nitratireducens* DSM 15102 *

Final pH: 7.1 - 7.4

Final volume: 1003 ml

Solution A	942.00	ml
Solution B	30.00	ml
Solution C	20.00	ml
Solution D	1.00	ml
Solution E	10.00	ml

1. Solution A is sparged with 80% N₂ and 20% CO₂ gas mixture to reach a pH below 6 (at least 30 min), then distributed under the same gas atmosphere in anoxic Hungate-type tubes or serum vials and autoclaved. Solution B is autoclaved separately under 80% N₂ and 20% CO₂ gas atmosphere. Solutions C and E are autoclaved under 100% N₂ gas. Solution D is prepared under 100% N₂ gas atmosphere and sterilized by filtration. To complete the medium appropriate amounts of solutions B to E are added to the sterile solution A in the sequence as indicated. Final pH of the medium should be 7.1 - 7.4.

2. Note: Addition of 10 - 20 mg sodium dithionite per liter (e.g. from 5% (w/v) solution, freshly prepared under N₂ and filter-sterilized) may stimulate growth of some strains at the beginning. For transfers use 5 - 10% (v/v) inoculum.

* Omit Na-acetate and add after autoclaving 1.00 g/l yeast extract and 3.50 g/l D-glucose from sterile anoxic stock solutions.

Solution A

Na ₂ SO ₄	3.00	g
KH ₂ PO ₄	0.20	g
NH ₄ Cl	0.30	g
NaCl	7.00	g
MgCl ₂ x 6 H ₂ O	1.30	g
KCl	0.50	g
CaCl ₂ x 2 H ₂ O	0.15	g
Trace element solution SL-10	1.00	ml
Selenite-tungstate solution	1.00	ml
Sodium resazurin (0.1% w/v)	0.50	ml
Yeast extract	1.00	g
Distilled water	940.00	ml

Solution B

Na ₂ CO ₃	1.50	g
---------------------------------	------	---

193: DESULFOBACTER MEDIUM

Distilled water	30.00	ml
-----------------	-------	----

Solution C

Na acetate x 3 H₂O	2.50	g
D-Glucose	3.50	g
Distilled water	20.00	ml

Solution D

Wolin's vitamin solution (10x)	1.00	ml
---------------------------------------	------	----

Solution E

Na ₂ S x 9 H ₂ O	0.40	g
Distilled water	10.00	ml

Selenite-tungstate solution (from medium 385)

NaOH	0.50	g
Na ₂ SeO ₃ x 5 H ₂ O	3.00	mg
Na ₂ WO ₄ x 2 H ₂ O	4.00	mg
Distilled water	1000.00	ml

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

First dissolve FeCl₂ in the HCl, then dilute in water, add and dissolve the other salts. Finally make up to 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

193: DESULFOBACTER MEDIUM

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