

## **198: DESULFOSARCINA MEDIUM (BRACKISH WATER)**

Final pH: 7.1 - 7.4 Final volume: 1003 ml

Solution A	952.00	ml
Solution B	30.00	ml
Solution C	10.00	ml
Solution D	1.00	ml
Solution E	10.00	ml

1. Solution A is sparged with 80% N<sub>2</sub> and 20% CO<sub>2</sub> gas mixture to reach a pH below 6 (at least 30 min), then distributed under the same gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclaved. Solution B is autoclaved separately under 80% N<sub>2</sub> and 20%  $CO_2$  gas atmosphere. Solutions C and E are autoclaved under 100%  $N_2$  gas. Solution D is prepared under 100%  $N_2$  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 N<sub>2</sub> and filter-sterilized) may stimulate growth of some strains at the beginning. For transfers use 5 - 10% (v/v) inoculum.

For DSM 2650: Na-benzoate is replaced by 1.50 g/l Na-pyruvate and 1.00 g/l Na<sub>2</sub>-malate added after autoclaving from sterile anoxic stock solutions prepared under N<sub>2</sub>.

For DSM 9990: Na-benzoate is replaced by 6.20 g/l Na-acetate.

For <u>DSM 103834</u>: Na-benzoate is replaced by 2.5 g/l Na-lactate.

For <u>DSM 113909</u>: Supplement medium with 1.60 g Na<sub>2</sub>-fumarate. Omit Na-benzoate. Adjust pH of final medium to 8.0 - 8.5.

For <u>DSM 115219</u>: Supplement medium with 0.35 g Na-formate and 0.16 g Na-acetate. Omit Na-benzoate.

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Solution A	
$Na_2SO_4$	
KH-PO.	

Na <sub>2</sub> SO <sub>4</sub>	3.00	g
KH <sub>2</sub> PO <sub>4</sub>	0.20	g
NH <sub>4</sub> Cl	0.30	g
NaCl	13.50	g
$MgCl_2 \times 6 H_2O$	2.00	g
KCI	0.50	g
$CaCl_2 \times 2 H_2O$	0.15	g
Selenite-tungstate solution	1.00	ml
Trace element solution SL-10	1.00	ml

## Microorganisms



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Sodium resazurin (0.1% w/v) Distilled water	0.50 950.00	ml ml
Solution B Na <sub>2</sub> CO <sub>3</sub> Distilled water	1.50 30.00	g
Distilled water	30.00	ml
Solution C		
Na-benzoate	0.60	g
Distilled water	10.00	ml
Solution D		
Wolin's vitamin solution (10x)	1.00	ml
Solution E		
Na <sub>2</sub> S x 9 $H_2O$	0.40	g
Distilled water	10.00	ml
Selenite-tungstate solution (from medium 3	385)	
NaOH	0.50	g
$Na_2SeO_3 \times 5 H_2O$	3.00	mg
$Na_2WO_4 \ge H_2O$	4.00	mg
Distilled water	1000.00	ml
Trace element solution SL-10 (from mediu	m 320)	
HCI (25%)	10.00	ml
$FeCl_2 \times 4 H_2O$	1.50	g
ZnCl <sub>2</sub>	70.00	mg
$MnCl_2 \times 4 H_2O$	100.00	mg
H <sub>3</sub> BO <sub>3</sub>	6.00	mg
$CoCl_2 \times 6 H_2O$	190.00	mg
$CuCl_2 \times 2 H_2O$	2.00	mg
$NiCl_2 \times 6 H_2O$	24.00	mg
$Na_2MoO_4 \times 2 H_2O$	36.00	mg
Distilled water	990.00	ml

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

## Microorganisms



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Wolin's vitamin solution (10x) (from medium 120)			
Biotin	20.00	mg	
Folic acid	20.00	mg	
Pyridoxine hydrochloride	100.00	mg	
Thiamine HCI	50.00	mg	
Riboflavin	50.00	mg	
Nicotinic acid	50.00	mg	
Calcium D-(+)-pantothenate	50.00	mg	
Vitamin B <sub>12</sub>	1.00	mg	
p-Aminobenzoic acid	50.00	mg	
(DL)-alpha-Lipoic acid	50.00	mg	
Distilled water	1000.00	ml	