

## 193: DESULFOBACTER MEDIUM

<b>Solution A</b>	942.00	ml
<b>Solution B</b>	30.00	ml
<b>Solution C</b>	20.00	ml
<b>Solution D</b>	1.00	ml
<b>Solution E</b>	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 in anoxic Hungate-type tubes or serum vials and autoclaved. Solution B is autoclaved separately under 80% N<sub>2</sub> and 20% CO<sub>2</sub> gas atmosphere. Solutions C and E are autoclaved under 100% N<sub>2</sub> gas. Solution D is prepared under 100% N<sub>2</sub> 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 1744](#), [DSM 10631](#), [DSM 16697](#): Omit Na-acetate and add after autoclaving 1.00 g/l yeast extract and 2.50 g/l Na-L-Lactate from sterile anoxic stock solutions.

For [DSM 2059](#), [DSM 27197](#): Omit Na-acetate and add after autoclaving 0.60 g/l Na-benzoate from an anoxic stock solution sterilized by filtration.

For [DSM 4660](#): Omit Na-acetate and add after autoclaving 1.00 ml/l seven vitamins solution (see medium 503) and 0.10 g phenol from anoxic stock solutions sterilized by filtration.

For [DSM 6637](#): Replace Na-acetate with 0.58 g/l acetone. Supplement medium with 3.40 g/l KNO<sub>3</sub> and omit solution E. Add after autoclaving 5.00 g/l Na-pyruvate from an anoxic stock solution sterilized by filtration.

For [DSM 8775](#): Omit Na-acetate and add after autoclaving 2.50 g/l Na-pyruvate from an anoxic stock solution sterilized by filtration.

For [DSM 10711](#): Omit Na-acetate and add after autoclaving 1.00 ml/l seven vitamins solution (see medium 503), 2.50 g/l Na-pyruvate and 0.50 g/l yeast extract from anoxic stock solutions sterilized by filtration.

For [DSM 14956](#): Omit Na-acetate and add after autoclaving 1.60 g/l methanol from an anoxic stock solution sterilized by filtration.

For [DSM 15102](#): 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.

For [DSM 18843](#): Omit Na-acetate and add after autoclaving 1.00 ml/l seven vitamins solution (see medium 503) and 0.62 g/l Na-stearate from anoxic stock solutions sterilized

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by filtration.

For DSM 25524: Omit Na-acetate and add after autoclaving 1.00 ml/l seven vitamins solution (see medium 503) and 0.33 g/l Na-octanoate from anoxic stock solution sterilized by filtration.

### Solution A

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	7.00	g
MgCl <sub>2</sub> x 6 H <sub>2</sub> O	1.30	g
KCl	0.50	g
CaCl <sub>2</sub> x 2 H <sub>2</sub> O	0.15	g
<b>Trace element solution SL-10</b>	1.00	ml
<b>Selenite-tungstate solution</b>	1.00	ml
Sodium resazurin (0.1% w/v)	0.50	ml
Distilled water	940.00	ml

### Solution B

Na <sub>2</sub> CO <sub>3</sub>	1.50	g
Distilled water	30.00	ml

### Solution C

Na-acetate x 3 H <sub>2</sub> O	2.50	g
Distilled water	20.00	ml

### Solution D

<b>Wolin's vitamin solution (10x)</b>	1.00	ml
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### Solution E

Na <sub>2</sub> S x 9 H <sub>2</sub> O	0.40	g
Distilled water	10.00	ml

### Selenite-tungstate solution (from medium 385)

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

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### Trace element solution SL-10 (from medium 320)

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

First dissolve FeCl<sub>2</sub> 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
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