

## 517. DESULFOBACTERIUM OLEOVORANS MEDIUM

### Solution A:

KH <sub>2</sub> PO <sub>4</sub>	0.20	g
NH <sub>4</sub> Cl	0.25	g
Na <sub>2</sub> SO <sub>4</sub>	4.00	g
NaCl	20.00	g
MgCl <sub>2</sub> x 6 H <sub>2</sub> O	3.40	g
CaCl <sub>2</sub> x 2 H <sub>2</sub> O	0.25	g
Distilled water	920.00	ml

### Solution B:

Trace element solution SL-10 (see medium 320)	1.00	ml
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### Solution C:

Selenite-tungstate solution (see medium 385)	1.00	ml
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### Solution D:

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

### Solution E:

Vitamin solution (see medium 141)	10.00	ml
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### Solution F:

Vitamin B <sub>12</sub> solution (50 µg/ml)	1.00	ml
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### Solution G:

Stearic acid	0.36	g
2 N NaOH	0.63	ml
Distilled water	10.00	ml

Heat the suspension in a closed bottle (with 100% N<sub>2</sub> gas atmosphere in head space) in a boiling water bath. Shake until stearate has dissolved, then autoclave. Stored stearate solution has to be remelted before use.

### Solution H:

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

*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 same gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclaved.

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*Solutions B, C, G, and H are autoclaved separately under 100% N<sub>2</sub> gas. Solution D is autoclaved under 80% N<sub>2</sub> and 20% CO<sub>2</sub> gas atmosphere. Solutions E and F are prepared under 100% N<sub>2</sub> gas atmosphere and sterilized by filtration. To complete the medium appropriate amounts of solutions B to H are added to the sterile solution A in the sequence as indicated. Adjust pH of complete medium to 7.1 - 7.4, if necessary.*

*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% inoculum.*