

### 383: DESULFOBACTERIUM MEDIUM

This recipe contains strain-specific modifications for *Desulfofustis glycolicus* DSM 9705 \*

Final pH: 7.0 - 7.2

Final volume: 1003 ml

|                   |        |    |
|-------------------|--------|----|
| <b>Solution A</b> | 952.00 | ml |
| <b>Solution B</b> | 20.00  | 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 in anoxic cultivation vials and autoclaved under the same gas atmosphere. Solution B is autoclaved separately under 80% N<sub>2</sub> and 20% CO<sub>2</sub> gas atmosphere. Solutions C and D are prepared under 100% N<sub>2</sub> gas and filter-sterilized. Solution E is autoclaved under 100% N<sub>2</sub> gas. 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 7.0 - 7.2.

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% inoculum. Incubate all strains in the dark.

\* Replace pyruvate with 1.00 g/l Na-glycolate 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                                   | 21.00  | g  |
| MgCl <sub>2</sub> x 6 H <sub>2</sub> O | 3.00   | 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                        | 950.00 | ml |

#### Solution B

|                                 |       |    |
|---------------------------------|-------|----|
| Na <sub>2</sub> CO <sub>3</sub> | 1.00  | g  |
| Distilled water                 | 20.00 | ml |

## 383: DESULFOBACTERIUM MEDIUM

### Solution C

|                        |                 |              |
|------------------------|-----------------|--------------|
| <del>Na pyruvate</del> | <del>2.50</del> | <del>g</del> |
| Na-glycolate           | 1.00            | g            |
| Distilled water        | 20.00           | ml           |

### Solution D

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

### 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 |

### 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 |

## 383: DESULFOBACTERIUM MEDIUM

|                         |         |    |
|-------------------------|---------|----|
| 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 |