

## 640a: THERMOCLOSTRIDIUM CAENICOLA MEDIUM

|  |         |    |
|--|---------|----|
| NH <sub>4</sub> Cl   | 0.90    | g  |
| NaCl   | 0.90    | g  |
| MgCl <sub>2</sub> x 6 H <sub>2</sub> O                         | 0.40    | g  |
| KH <sub>2</sub> PO <sub>4</sub>                                | 0.75    | g  |
| K <sub>2</sub> HPO <sub>4</sub>                                | 1.50    | g  |
| Trypticase peptone (BD BBL)                                    | 2.00    | g  |
| Yeast extract (OXOID)  | 1.00    | g  |
| <b>Trace element solution SL-10</b>                            | 1.00    | ml |
| FeCl <sub>3</sub> x 6 H <sub>2</sub> O (0.1% w/v in 0.2 N HCl) | 2.50    | ml |
| Sodium resazurin (0.1% w/v)                                    | 0.50    | ml |
| NaHCO <sub>3</sub>   | 2.00    | g  |
| Cellobiose   | 1.00    | g  |
| L-Cysteine HCl x H <sub>2</sub> O                              | 0.50    | g  |
| Distilled water  | 1000.00 | ml |

Dissolve ingredients except bicarbonate, cellobiose and cysteine. Sparge medium with 80% N<sub>2</sub> and 20% CO<sub>2</sub> gas mixture for 30 - 45 min to make it anoxic, then add bicarbonate and adjust pH to 6.8. Distribute medium under same gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. Add cellobiose and cysteine after autoclaving from anoxic stock solutions prepared under 100% N<sub>2</sub> gas atmosphere. Cellobiose has to be sterilized by filtration. Adjust pH of the complete medium to 6.5, if necessary.

### 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.