

213. SYNTHROPHOMONAS MEDIUM (SULFATE FREE)

Solution A:

| | | |
|---|--------|----|
| KH ₂ PO ₄ | 0.50 | g |
| MgCl ₂ x 6 H ₂ O | 0.33 | g |
| NaCl | 0.40 | g |
| NH ₄ Cl | 0.40 | g |
| CaCl ₂ x 2 H ₂ O | 0.05 | g |
| Trace element solution SL-10 (see medium 320) | 1.00 | ml |
| Rumen fluid, clarified (see medium 1310) | 50.00 | ml |
| Trypticase peptone (BD BBL) | 1.00 | g |
| Butyric acid | 1.70 | g |
| Na-resazurin solution (0.1% w/v) | 0.50 | ml |
| Distilled water | 850.00 | ml |

Solution B:

| | | |
|---------------------------------|-------|----|
| Na ₂ CO ₃ | 1.50 | g |
| Distilled water | 30.00 | ml |

Solution C:

| | | |
|--|------|----|
| Seven vitamins solution (see medium 503) | 1.00 | ml |
|--|------|----|

Solution D:

| | | |
|-----------------------------------|-------|----|
| L-Cysteine-HCl x H ₂ O | 0.30 | g |
| Distilled water | 10.00 | ml |

Solution E:

| | | |
|--|-------|----|
| Na ₂ S x 9 H ₂ O | 0.30 | g |
| Distilled water | 10.00 | ml |

Add and dissolve ingredients of *solution A*, adjust pH to 7.2 and sparge medium with 80% N₂ and 20% CO₂ gas mixture for 30 – 45 min to make it anoxic. Dispense medium under the same gas atmosphere into anoxic Hungate-type tubes and autoclave. *Solution B* is prepared under 80% N₂ and 20% CO₂ gas atmosphere and autoclaved. *Solution C* is prepared under 100% N₂ gas and sterilized by filtration. *Solutions D* and *E* are autoclaved under 100% N₂ gas atmosphere. To complete the medium appropriate amounts of *solutions B* to *E* are added to the sterile *solution A* in the sequence as indicated.

Continued next page

Note: Some cultures are shipped in semi-solid medium which stimulates growth at the beginning. For agar stabs 3.00 g/l agar are added to the complete medium from a sterile anoxic stock solution (2% w/v). Upon receipt add anoxically 1 - 2 ml of the recommended freshly prepared liquid medium to the agar tube and incubate for 3 - 5 days. After incubation transfer 0.5 ml of the resulting cell suspension in the liquid phase to tubes with liquid medium.

For [DSM 2909](#), [DSM 2984](#) and [DSM 21899](#) replace butyric acid with 1.50 g/l acetoin.

For [DSM 15682](#) and [DSM 16215](#) replace butyric acid with 1.70 g/l crotonic acid.

For [DSM 102353](#) replace butyric acid with 2.00 g/l Na-benzoate.