1166a: DESULFURIBACILLUS MEDIUM (ARSENATE)

NaCl 6.00 g
K₂HPO₄ 1.00 g
Na₂SO₄ 2.80 g
NaHCO₃ 8.00 g
Na₂CO₃ 22.00 g
NH₄Cl 0.20 g
MgCl₂ x 6 H₂O 0.20 g
Trace elements solution (Pfennig & Lippert, 1966) 1.00 ml
Selenite-tungstate solution 1.00 ml
Yeast extract 0.05 g
Na-pyruvate 2.20 g
Na₂H₂AsO₄ x 7 H₂O 6.20 g
Wolin’s vitamin solution (10x) 1.00 ml
Na₂S x 9 H₂O 0.24 g
Distilled water 1000.00 ml

Dissolve sodium chloride, hydrogenphosphate and sodium sulfate, then sparge solution with 100% N₂ gas for 30 - 45 min to make it anoxic. Add and dissolve hydrogencarbonate and carbonate, then dispense under 100% N₂ gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. After sterilization add ammonium chloride, magnesium chloride, trace elements, yeast extract, pyruvate, arsenate, vitamins and sulfide from sterile anoxic stock solutions prepared under 100% N₂ gas. Stock solutions of vitamins, pyruvate and arsenate should be sterilized by filtration. Adjust pH of complete medium to 9.5 - 10.

Trace elements solution (Pfennig & Lippert, 1966) (from medium 1369)
EDTA 5.00 g
FeSO₄ x 7 H₂O 2.20 g
ZnSO₄ x 7 H₂O 0.10 g
MnCl₂ x 4 H₂O 0.03 g
H₃BO₃ 0.03 g
CoCl₂ x 6 H₂O 0.20 g
CuCl₂ x 2 H₂O 0.03 g
NiCl₂ x 6 H₂O 0.03 g
Na₂MoO₄ x 2 H₂O 0.03 g
Distilled water 1000.00 ml

pH 3.0-4.0
### Selenite-tungstate solution (from medium 385)

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>Quantity</th>
</tr>
</thead>
<tbody>
<tr>
<td>NaOH</td>
<td>0.50 g</td>
</tr>
<tr>
<td>Na₂SeO₃ x 5 H₂O</td>
<td>3.00 mg</td>
</tr>
<tr>
<td>Na₂WO₄ x 2 H₂O</td>
<td>4.00 mg</td>
</tr>
<tr>
<td>Distilled water</td>
<td>1000.00 ml</td>
</tr>
</tbody>
</table>

### Wolin's vitamin solution (10x) (from medium 120)

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>Quantity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Biotin</td>
<td>20.00 mg</td>
</tr>
<tr>
<td>Folic acid</td>
<td>20.00 mg</td>
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<tr>
<td>Pyridoxine hydrochloride</td>
<td>100.00 mg</td>
</tr>
<tr>
<td>Thiamine HCl</td>
<td>50.00 mg</td>
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<tr>
<td>Riboflavin</td>
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<tr>
<td>Nicotinic acid</td>
<td>50.00 mg</td>
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<tr>
<td>Calcium D-(+)-pantothenate</td>
<td>50.00 mg</td>
</tr>
<tr>
<td>Vitamin B₁₂</td>
<td>1.00 mg</td>
</tr>
<tr>
<td>p-Aminobenzoic acid</td>
<td>50.00 mg</td>
</tr>
<tr>
<td>(DL)-alpha-Lipoic acid</td>
<td>50.00 mg</td>
</tr>
<tr>
<td>Distilled water</td>
<td>1000.00 ml</td>
</tr>
</tbody>
</table>