1115: CONGREGIBACTER (SYPHC) MEDIUM

Sea Salt (SIGMA) 35.00 g
**Wolfe’s mineral elixir** 1.00 ml
NH₄Cl 0.10 g
Yeast extract 1.00 g
Na-pyruvate 1.10 g
HEPES (SIGMA) 2.50 g
L-Histidine 0.04 g
L-Cysteine HCl x H₂O 0.04 g
**Seven vitamins solution** 0.50 ml
KH₂PO₄ 0.05 g
Distilled water 1000.00 ml

1. Dissolve ingredients (except vitamins and hydrogenphosphate) and adjust pH to 7.7. After autoclaving add vitamins and hydrogenphosphate from sterile stock solutions sterilized by filtration. Adjust pH of complete medium to 7.3 - 7.5, if necessary.

2. Note: Growth of most strains is stimulated by using microaerophilic conditions with only 10% (v/v) oxygen in the head space gas atmosphere. For microaerophilic growth medium is prepared without HEPES, sparged with 80% N₂ and 20% CO₂ gas mixture to make it anoxic and then dispensed under air atmosphere into serum vials to 50% volume. After autoclaving the pH is adjusted to 7.5 using a sterile anoxic stock solution of Na₂CO₃ (5% w/v) prepared under 80% N₂ and 20% CO₂ gas mixture.

**Wolfe’s mineral elixir** (from medium 792)

MgSO₄ x 7 H₂O 30.00 g
MnSO₄ x H₂O 5.00 g
NaCl 10.00 g
FeSO₄ x 7 H₂O 1.00 g
CoCl₂ x 6 H₂O 1.80 g
CaCl₂ x 2 H₂O 1.00 g
ZnSO₄ x 7 H₂O 1.80 g
CuSO₄ x 5 H₂O 0.10 g
AlK(SO₄)₂ x 12 H₂O 0.18 g
H₃BO₃ 0.10 g
Na₂MoO₄ x 2 H₂O 0.10 g
(NH₄)₂Ni(SO₄)₂ x 6 H₂O 2.80 g
Na₂WO₄ x 2 H₂O 0.10 g
Na₂SeO₄ 0.10 g
Distilled water 1000.00 ml

First adjust pH to 1.0 with diluted H₂SO₄, then add and dissolve the salts.
### Seven vitamins solution (from medium 503)

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>Amount</th>
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<tbody>
<tr>
<td>Vitamin B&lt;sub&gt;12&lt;/sub&gt;</td>
<td>100.00 mg</td>
</tr>
<tr>
<td>p-Aminobenzoic acid</td>
<td>80.00 mg</td>
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<tr>
<td>D-(+)-biotin</td>
<td>20.00 mg</td>
</tr>
<tr>
<td>Nicotinic acid</td>
<td>200.00 mg</td>
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<tr>
<td>Calcium pantothenate</td>
<td>100.00 mg</td>
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<tr>
<td>Pyridoxine hydrochloride</td>
<td>300.00 mg</td>
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<tr>
<td>Thiamine-HCl x 2 H&lt;sub&gt;2&lt;/sub&gt;O</td>
<td>200.00 mg</td>
</tr>
<tr>
<td>Distilled water</td>
<td>1000.00 ml</td>
</tr>
</tbody>
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