

## 81a: THIOMONAS ISLANDICA MEDIUM

K <sub>2</sub> HPO <sub>4</sub>	2.30	g
Na <sub>2</sub> HPO <sub>4</sub> x 2 H <sub>2</sub> O	2.90	g
NH <sub>4</sub> Cl	1.00	g
CaCl <sub>2</sub> x 2 H <sub>2</sub> O	0.01	g
MgSO <sub>4</sub> x 7 H <sub>2</sub> O	0.50	g
NaVO <sub>3</sub> x H <sub>2</sub> O (0.1% w/v)	5.00	ml
<b>Trace element solution SL-6</b>	5.00	ml
NaHCO <sub>3</sub>	0.50	g
Ferric ammonium citrate	0.05	g
<b>Seven vitamins solution</b>	1.00	ml
Distilled water	1000.00	ml

1. Dissolve ingredients (except bicarbonate, ferric ammonium citrate and vitamins), then sparge solution with 80% N<sub>2</sub> and 20% CO<sub>2</sub> gas mixture for at least 30 - 45 min to remove dissolved oxygen and to saturate the solution with CO<sub>2</sub>. Adjust pH to 4.5 and dispense solution under air atmosphere into serum vials suitable for anaerobic cultivation to 50% of volume. Close vials with butyl rubber septa to prevent free exchange of oxygen with the external atmosphere and autoclave. Add ferric ammonium citrate and vitamins from anoxic stock solutions sterilized by filtration and bicarbonate from a sterile stock solution prepared under 80% N<sub>2</sub> and 20% CO<sub>2</sub> gas atmosphere. Adjust pH of the complete medium to 5.0, if necessary.
2. After inoculation add sterile 80% H<sub>2</sub> and 20% CO<sub>2</sub> gas mixture to 2 bar overpressure.
3. Note: Use at least 10% (v/v) as inoculum.

### Trace element solution SL-6 (from medium 27)

ZnSO <sub>4</sub> x 7 H <sub>2</sub> O	0.10	g
MnCl <sub>2</sub> x 4 H <sub>2</sub> O	0.03	g
H <sub>3</sub> BO <sub>3</sub>	0.30	g
CoCl <sub>2</sub> x 6 H <sub>2</sub> O	0.20	g
CuCl <sub>2</sub> x 2 H <sub>2</sub> O	0.01	g
NiCl <sub>2</sub> x 6 H <sub>2</sub> O	0.02	g
Na <sub>2</sub> MoO <sub>4</sub> x 2 H <sub>2</sub> O	0.03	g
Distilled water	1000.00	ml

### Seven vitamins solution (from medium 503)

Vitamin B <sub>12</sub>	100.00	mg
p-Aminobenzoic acid	80.00	mg
D-(+)-biotin	20.00	mg



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Nicotinic acid	200.00	mg
Calcium pantothenate	100.00	mg
Pyridoxine hydrochloride	300.00	mg
Thiamine-HCl x 2 H <sub>2</sub> O	200.00	mg
Distilled water	1000.00	ml