

## 1166: DESULFONATRONUM THIOAUTOTROPHICUM MEDIUM

This recipe contains strain-specific modifications for *Natranaerovirga hydrolytica* DSM 24176 \*

Final pH: 9.5 - 10.0

Final volume: 1003 ml

NaCl	6.00	g
K <sub>2</sub> HPO <sub>4</sub>	1.00	g
Na <sub>2</sub> SO <sub>4</sub>	2.80	g
NaHCO <sub>3</sub>	8.00	g
Na <sub>2</sub> CO <sub>3</sub>	22.00	g
NH <sub>4</sub> Cl	0.20	g
MgCl <sub>2</sub> x 6 H <sub>2</sub> O	0.20	g
<b>Trace elements solution (Pfennig &amp; Lippert,1966)</b>	1.00	ml
<b>Selenite-tungstate solution</b>	1.00	ml
Yeast extract	0.05	g
<del>Na formate</del>	<del>3.40</del>	<del>g</del>
<del>Na pyruvate</del>	<del>0.55</del>	<del>g</del>
Na <sub>2</sub> S <sub>2</sub> O <sub>3</sub> x 5 H <sub>2</sub> O	5.00	g
<b>Wolin's vitamin solution (10x)</b>	1.00	ml
Na <sub>2</sub> S x 9 H <sub>2</sub> O	0.24	g
D-galacturonic acid	1.00	g
Distilled water	1000.00	ml

Dissolve sodium chloride, hydrogenphosphate and sodium sulfate, then sparge solution with 100% N<sub>2</sub> gas for 30 - 45 min to make it anoxic. Add and dissolve carbonates, dispense under 100% N<sub>2</sub> gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. After sterilization add ammonium chloride, magnesium chloride, trace elements, yeast extract, formate, pyruvate, thiosulfate, vitamins and sulfide from sterile anoxic stock solutions prepared under 100% N<sub>2</sub> gas. Stock solutions of vitamins and thiosulfate should be sterilized by filtration. Adjust pH of complete medium to 9.5 - 10.

\* Replace formate and pyruvate with 1.00 g/l D-galacturonic acid sodium salt as substrate, added after autoclaving from an anoxic stock solution sterilized by filtration.

### Trace elements solution (Pfennig & Lippert,1966) (from medium 1369)

EDTA	5.00	g
FeSO <sub>4</sub> x 7 H <sub>2</sub> O	2.20	g
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>	0.03	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	0.03	g

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NiCl <sub>2</sub> x 6 H <sub>2</sub> O	0.03	g
Na <sub>2</sub> MoO <sub>4</sub> x 2 H <sub>2</sub> O	0.03	g
Distilled water	1000.00	ml

pH 3.0-4.0

### **Selenite-tungstate solution** (from medium 385)

NaOH	0.50	g
Na <sub>2</sub> SeO <sub>3</sub> x 5 H <sub>2</sub> O	3.00	mg
Na <sub>2</sub> WO <sub>4</sub> x 2 H <sub>2</sub> O	4.00	mg
Distilled water	1000.00	ml

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

Biotin	20.00	mg
Folic acid	20.00	mg
Pyridoxine hydrochloride	100.00	mg
Thiamine HCl	50.00	mg
Riboflavin	50.00	mg
Nicotinic acid	50.00	mg
Calcium D-(+)-pantothenate	50.00	mg
Vitamin B <sub>12</sub>	1.00	mg
p-Aminobenzoic acid	50.00	mg
(DL)-alpha-Lipoic acid	50.00	mg
Distilled water	1000.00	ml