## **Microorganisms**



#### **541: SULFUROSPIRILLUM MEDIUM**

Solution A	952.00	ml
Solution B	30.00	ml
Solution C	20.00	ml
Solution D	2.00	ml

Sparge solution A with 80%  $N_2$  and 20%  $CO_2$  gas mixture for 30 – 45 min to make it anoxic, distribute under same gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. Solution B is autoclaved separately under 80%  $N_2$  and 20%  $CO_2$  gas atmosphere. Solution C should be prepared under 100%  $N_2$  gas and sterilized by filtration. Solution D is autoclaved under 100%  $N_2$  gas atmosphere. To complete the medium appropriate amounts of solutions B to D are added to the sterile solution A in the sequence as indicated. Adjust pH of complete medium to 7.2, if necessary.

For <u>DSM 18149</u>: Supplement medium after autoclaving with 1.00 ml/l Wolin's vitamin solution 10x (see medium 120) sterilized by filtration. Prior to inoculation reduce the complete medium with 10 - 20 mg/l sodium dithionite (added from a 5% w/v stock solution freshly prepared under 100% N<sub>2</sub> gas and filter-sterilized) instead of using cysteine.

For <u>DSM 22742</u>: Supplement medium with 1.00 ml/l Wolin's vitamin solution 10x (see medium 120) and 0.50 g/l Na-thiosulfate added after autoclaving from anoxic stock solutions sterilized by filtration.

#### Solution A

KH <sub>2</sub> PO <sub>4</sub>	1.36	g
$MgSO_4 \times 7 H_2O$	0.37	g
CaCl <sub>2</sub> x 2 H <sub>2</sub> O	0.10	g
NH <sub>4</sub> Cl	0.27	g
Trace element solution SL-10	2.00	ml
Distilled water	950.00	ml

### **Solution B**

Na <sub>2</sub> CO <sub>3</sub>	1.50	g
Distilled water	30.00	ml

#### **Solution C**

Na <sub>2</sub> -fumarate	4.00	g
Distilled water	20.00	ml

#### Solution D

L-Cysteine HCl x H <sub>2</sub> O	0.07	а

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#### **541: SULFUROSPIRILLUM MEDIUM**



Distilled water	2.00	ml
Distinca water	2.00	

### Trace element solution SL-10 (from medium 320)

HCI (25%)	10.00	ml
FeCl <sub>2</sub> x 4 H <sub>2</sub> O	1.50	g
ZnCl <sub>2</sub>	70.00	mg
$MnCl_2 \times 4 H_2O$	100.00	mg
$H_3BO_3$	6.00	mg
CoCl <sub>2</sub> x 6 H <sub>2</sub> O	190.00	mg
CuCl <sub>2</sub> x 2 H <sub>2</sub> O	2.00	mg
$NiCl_2 \times 6 H_2O$	24.00	mg
$Na_2MoO_4 \times 2 H_2O$	36.00	mg
Distilled water	990.00	ml

First dissolve  $\text{FeCl}_2$  in the HCl, then dilute in water, add and dissolve the other salts. Finally make up to 1000.00 ml.