# **Microorganisms**



### 838: GEOANAEROBACTER MEDIUM

Fe(III) citrate (19% Fe)	10.00	g
KH <sub>2</sub> PO <sub>4</sub>	0.60	g
NH <sub>4</sub> Cl	0.30	g
$MgSO_4 \times 7 H_2O$	0.50	g
CaCl <sub>2</sub> x 2 H <sub>2</sub> O	0.10	g
Trace element solution SL-11	1.00	ml
Selenite-tungstate solution	1.00	ml
Na <sub>2</sub> CO <sub>3</sub>	1.50	g
Na-acetate	0.80	g
Na-ascorbate	0.80	g
Distilled water	1000.00	ml

First dissolve ferric citrate by heating the water under continuous stirring. After cooling to room temperature adjust the pH to 6.0, then add and dissolve the remaining ingredients, except carbonate, acetate and ascorbate. Sparge medium with 80%  $N_2$  and 20%  $CO_2$  gas mixture for 30 - 45 min to make it anoxic, then dispense under the same gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. After autoclaving add acetate and ascorbate from sterile anoxic stock solutions prepared under 100%  $N_2$  gas and carbonate from a sterile anoxic stock solution prepared under 80%  $N_2$  and 20%  $CO_2$  gas mixture. Prior to inoculation check the medium pH and adjust to 6.8, if necessary.

For <u>DSM 9736</u>: Supplement medium with 1.00 g/l NaCl. After autoclaving supplement medium with 1.00 ml/l of seven vitamins solution (see medium 503) and replace Na -ascorbate with 0.15 g/ of L-cysteine-HCl x  $H_2O$  added from a sterile anoxic stock solution prepared under 100%  $N_2$  gas.

For <u>DSM 15288</u>, <u>DSM 16228</u>: Omit Na-acetate and Na-ascorbate. Supplement medium after autoclaving with 10.00 ml/l of a Wolin's vitamin solution (see medium 141), 1.00 g/l Na-DL-lactate and 1.00 g/l yeast extract added from sterile anoxic stock solutions prepared under 100%  $N_2$  gas.

For <u>DSM 16401</u>: Supplement medium with 20.00 g/l NaCl and omit Na-ascorbate. After autoclaving supplement medium with 10.00 ml/l of a sterile anoxic Wolin's vitamin solution (see medium 141).

For <u>DSM 19350</u>, <u>DSM 22248</u>, <u>DSM 24905</u>, <u>DSM 26175</u>: Prepare medium without Na -ascorbate.

### Trace element solution SL-11 (from medium 722)

$Na_2$ -EDTA x 2 $H_2O$	5.20	g
FeCl <sub>2</sub> x 4 H <sub>2</sub> O	1.50	g
ZnCl <sub>2</sub>	70.00	mg
MnCl <sub>2</sub> x 4 H <sub>2</sub> O	100.00	mg

# **Microorganisms**

### 838: GEOANAEROBACTER MEDIUM



H <sub>3</sub> BO <sub>3</sub>	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	1000.00	ml

Dissolve EDTA in 800 ml distilled water, adjust pH to 7 using 2 N NaOH and add ferrous chloride. After ferrous chloride has dissolved add other compounds. Finally adjust pH to 6.0 and bring volume to 1000 ml.

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

NaOH 0.50	) g
$Na_2SeO_3 \times 5 H_2O$ 3.00	) mg
$Na_2WO_4 \times 2 H_2O$ 4.00	) mg
Distilled water 1000.00	) ml