## **Microorganisms**



## 113: THIOBACILLUS DENITRIFICANS MEDIUM

Final pH: 7.0 - 7.2 Final volume: 1003 ml

Solution A	962.00	ml
Solution B	20.00	ml
Solution C	20.00	ml
Solution D	1.00	ml

Solutions A, B, and D are sterilized separately by autoclaving at  $121^{\circ}$ C for 15 min under a 100% N<sub>2</sub> gas atmosphere. Solution C is sterilized by filtration under an atmosphere of 80% N<sub>2</sub> and 20% CO<sub>2</sub> gas mixture. Appropriate amounts of solutions B to D are added to the sterile solution A in the sequence as indicated. Check the pH of the complete medium and adjust it to 7.0 - 7.2, if necessary.

Solution A		
$KH_2PO_4$	2.00	g
KNO <sub>3</sub>	2.00	g
NH <sub>4</sub> Cl	1.00	g
$MgSO_4 \times 7 H_2O$	0.80	g
Trace element solution SL-4	2.00	ml
Distilled water	960.00	ml
Solution B		
$Na_2S_2O_3 \times 5 H_2O$	5.00	g
Distilled water	20.00	ml
Solution C	1.00	
Na <sub>2</sub> CO <sub>3</sub>		g
Distilled water	20.00	ml
Solution D	2.00	
FeSO <sub>4</sub> x 7 H <sub>2</sub> O	2.00	mg
H <sub>2</sub> SO <sub>4</sub> (0.1 N)	1.00	ml
Trace element solution SL-4 (from medium 14)		_
Na <sub>2</sub> -EDTA	0.50	g
$FeSO_4 \times 7 H_2O$	0.20	g

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$ZnSO_4 \times 7 H_2O$	0.10	g
$MnCl_2 \times 4 H_2O$	0.03	g
$H_3BO_3$	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
$Na_2MoO_4 \times 2 H_2O$	0.03	g
Distilled water	1000.00	ml

First dissolve EDTA in distilled water and adjust pH to 7.0 using 2 N NaOH; then add other compounds.