

1801: SULFURIMONAS MARISNIGRI MEDIUM

Solution A	1001.00	ml
Solution B	15.00	ml
Solution C	2.00	ml
Solution D	2.00	ml
Solution E	2.00	ml
Solution F	3.00	ml
Solution G	1.00	ml

Dissolve ingredients of solution A (except MnO_2), then sparge with 80% N_2 and 20% CO_2 gas mixture for 30 - 45 min to make it anoxic. Distribute solution A under the same gas atmosphere into anoxic Hungate-type tubes or serum vials containing the appropriate amount of MnO_2 and autoclave at 121°C for 15 min. Solution B is autoclaved separately under an 80% N_2 and 20% CO_2 gas atmosphere, and solutions C, D, and E are autoclaved under a 100% N_2 gas atmosphere. Solutions F and G are prepared under 100% N_2 gas atmosphere and sterilized by filtration. To complete the medium appropriate amounts of solutions B to G are added to the sterile solution A in the sequence as indicated. The final pH of the complete medium should be 7.5.

Solution A

NaCl	17.70	g
$\text{MgCl}_2 \times 6 \text{H}_2\text{O}$	3.20	g
$\text{CaCl}_2 \times 2 \text{H}_2\text{O}$	0.40	g
KCl	0.70	g
$\text{MgSO}_4 \times \text{H}_2\text{O}$	2.70	g
Trace element solution SL-10	1.00	ml
MnO_2	0.50	g
Distilled water	1000.00	ml

Solution B

Na_2CO_3	0.75	g
Distilled water	15.00	ml

Solution C

NH_4Cl	0.01	g
Distilled water	2.00	ml

Solution D

K_2HPO_4	0.01	g
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Distilled water	2.00	ml
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Solution E

Na-acetate	0.16	g
Distilled water	2.00	ml

Solution F

$\text{Na}_2\text{S}_2\text{O}_3 \times 5 \text{H}_2\text{O}$	0.75	g
Distilled water	3.00	ml

Solution G

Wolin's vitamin solution (10x)	1.00	ml
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Trace element solution SL-10 (from medium 320)

HCl (25%)	10.00	ml
$\text{FeCl}_2 \times 4 \text{H}_2\text{O}$	1.50	g
ZnCl_2	70.00	mg
$\text{MnCl}_2 \times 4 \text{H}_2\text{O}$	100.00	mg
H_3BO_3	6.00	mg
$\text{CoCl}_2 \times 6 \text{H}_2\text{O}$	190.00	mg
$\text{CuCl}_2 \times 2 \text{H}_2\text{O}$	2.00	mg
$\text{NiCl}_2 \times 6 \text{H}_2\text{O}$	24.00	mg
$\text{Na}_2\text{MoO}_4 \times 2 \text{H}_2\text{O}$	36.00	mg
Distilled water	990.00	ml

First dissolve FeCl_2 in the HCl, then dilute in water, add and dissolve the other salts. Finally make up to 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_{12}	1.00	mg
p-Aminobenzoic acid	50.00	mg
(DL)-alpha-Lipoic acid	50.00	mg
Distilled water	1000.00	ml