

311c: SPOROMUSA ACIDOVORANS MEDIUM

NH ₄ Cl	0.50	g
MgSO ₄ x 7 H ₂ O	0.50	g
CaCl ₂ x 2 H ₂ O	0.25	g
NaCl	2.25	g
FeSO ₄ x 7 H ₂ O (0.1% w/v in 0.1 N H ₂ SO ₄)	2.00	ml
Trace element solution SL-10	1.00	ml
Selenite-tungstate solution	1.00	ml
Yeast extract	2.00	g
Casitone	2.00	g
Sodium resazurin (0.1% w/v)	0.50	ml
K ₂ HPO ₄	0.35	g
KH ₂ PO ₄	0.23	g
Na ₂ CO ₃	1.00	g
D-Fructose	5.00	g
Wolin's vitamin solution (10x)	1.00	ml
L-Cysteine HCl x H ₂ O	0.30	g
Na ₂ S x 9 H ₂ O	0.30	g
Distilled water	1000.00	ml

Dissolve ingredients (except phosphates, carbonate, fructose, vitamins, cysteine and sulfide) and sparge medium with 80% N₂ and 20% CO₂ gas mixture for 30 - 45 min to make it anoxic. Dispense medium under same gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. After autoclaving add phosphates, fructose, vitamins, cysteine and sulfide to the medium from sterile anoxic stock solutions prepared under 100% N₂ gas and carbonate from a sterile anoxic stock solution prepared under 80% N₂ and 20% CO₂ gas mixture. Stock solutions of fructose and vitamins should be sterilized by filtration. Adjust pH of complete medium to pH 6.5 - 7.0, if necessary.

For DSM 4440: Replace fructose with 1.35 g/l betaine as substrate; cysteine and sulfide must be replaced by 0.15 g/l DL-dithiothreitol (DTT) added from an anoxic stock solution sterilized by filtration.

For DSM 6539, DSM 6540, DSM 17108: Replace fructose with 2.00 g/l D-glucose added to the autoclaved medium from a filter-sterilized anoxic stock solution. Cysteine and sulfide must be replaced by 0.15 g/l DL-dithiothreitol (DTT) added from an anoxic stock solution sterilized by filtration.

For DSM 11501: Supplement medium with 0.30 g DL-Dithiothreitol and 20 ml/l methanol (50% v/v). Omit D-fructose, Na₂S x 9 H₂O, and L-Cysteine HCl x H₂O. After inoculation, add 1 bar overpressure of sterile 80% H₂ and 20% CO₂ gas mixture.

For DSM 14980: Replace fructose with 8.00 g/l D-glucose and 2.00 g/l Na-pyruvate as substrates added to the autoclaved medium from filter-sterilized anoxic stock solutions.

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For DSM 16652: Replace fructose with 1.11 g/l N-acetyl-D-glucosamine as substrate added to the autoclaved medium from a filter-sterilized anoxic stock solution.

For DSM 17189, DSM 17285: Replace fructose with 3.60 g/l D-mannitol as substrate added to the autoclaved medium from a filter-sterilized anoxic stock solution. Cysteine and sulfide must be replaced by 0.30 g/l DL-dithiothreitol (DTT) added from an anoxic stock solution sterilized by filtration.

For DSM 26537: Replace fructose with 5.00 g/l D-glucose as substrate added to the autoclaved medium from a filter-sterilized anoxic stock solution. Adjust pH of completed medium to 7.8 using a sterile anoxic stock solution of 5% (w/v) Na_2CO_3 .

For DSM 26827: Replace fructose with 2.50 g/l Na-(DL)-lactate added to the autoclaved medium from a separately sterilized anoxic stock solution. Cysteine and sulfide must be replaced by 0.15 g/l DL-dithiothreitol (DTT) added from an anoxic stock solution sterilized by filtration.

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.

Selenite-tungstate solution (from medium 385)

NaOH	0.50	g
$\text{Na}_2\text{SeO}_3 \times 5 \text{ H}_2\text{O}$	3.00	mg
$\text{Na}_2\text{WO}_4 \times 2 \text{ H}_2\text{O}$	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



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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