

311a: SPOROMUSA MEDIUM (H₂/CO₂)

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
Wolin's vitamin solution (10x)	1.00	ml
DL-Dithiothreitol	0.15	g
Distilled water	1000.00	ml

Dissolve ingredients (except phosphates, carbonate, vitamins, and dithiothreitol) and sparge medium with 80% H₂ and 20% CO₂ gas mixture for 30 - 45 min to make it anoxic. Dispense under the same gas atmosphere in anoxic Hungate-type tubes or serum vials to 30% of their volume and autoclave. Add phosphates to the medium after autoclaving from sterile stock solutions prepared under 100% N₂ gas and carbonate from a sterile stock solution prepared under 80% N₂ and 20% CO₂ gas mixture. Vitamins and dithiothreitol should be prepared under N₂ gas and sterilized by filtration. Adjust the pH of the completed medium to pH 7.0.

For [DSM 11379](#), [DSM 11380](#), [DSM 11381](#), [DSM 11382](#): Supplement medium with 2.50 g/l Na-pyruvate.

Trace element solution SL-10 (from medium 320)

HCl (25%)	10.00	ml
FeCl ₂ x 4 H ₂ O	1.50	g
ZnCl ₂	70.00	mg
MnCl ₂ x 4 H ₂ O	100.00	mg
H ₃ BO ₃	6.00	mg
CoCl ₂ x 6 H ₂ O	190.00	mg
CuCl ₂ x 2 H ₂ O	2.00	mg
NiCl ₂ x 6 H ₂ O	24.00	mg
Na ₂ MoO ₄ x 2 H ₂ O	36.00	mg
Distilled water	990.00	ml

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First dissolve FeCl₂ 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
Na ₂ SeO ₃ x 5 H ₂ O	3.00	mg
Na ₂ WO ₄ x 2 H ₂ 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
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