Microorganisms



684: SYNTROPHOBACTER (MPOB) MEDIUM

$Na_2HPO_4 \times 2 H_2O$	0.53	g
KH ₂ PO ₄	0.41	g
NH ₄ Cl	0.30	g
CaCl ₂ x 2 H ₂ O	0.11	g
MgCl ₂ x 6 H ₂ O	0.10	g
NaCl	0.30	g
Trace element solution SL-10	1.00	ml
Selenite-tungstate solution	1.00	ml
Yeast extract	0.20	g
Sodium resazurin (0.1% w/v)	0.50	ml
Na_2CO_3	1.50	g
Na ₂ -fumarate	3.20	g
Wolin's vitamin solution (10x)	1.00	ml
$Na_2S \times 9 H_2O$	0.50	g
Distilled water	1000.00	ml

- 1. Dissolve ingredients (except carbonate, vitamins, fumarate and sulfide) and sparge medium with 80% N_2 and 20% CO_2 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 complete the medium by adding vitamins, fumarate and sulfide 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 atmosphere. Stock solutions of vitamins and fumarate are sterilized by filtration. Adjust pH of the complete medium to 7.0 7.2. After inoculation pressurize culture vials to 0.7 bar overpressure with sterile 80% N_2 and 20% CO_2 gas mixture.
- 2. Note: A white precipitate forms in this medium after autoclaving, which has however no negative effect on growth.

For <u>DSM 14055</u>: Replace Na_2 -fumarate with 2.50 g/l Na-pyruvate added after autoclaving from a sterile anoxic solution prepared under 100% N_2 gas atmosphere.

For <u>DSM 14823</u>: Replace Na_2 -fumarate with 5.00 g/l glucose. Sterilize glucose separately by filtration under 100% N_2 gas atmosphere. Reduce amount of Na_2CO_3 to 1.00 g/l and adjust pH to 6.5 - 6.8.

For DSM 17232: Replace Na_2 -fumarate with 2.00 ml/l methanol added after autoclaving from a sterile anoxic stock solution prepared under 100% N_2 gas atmosphere.

For DSM 23957: Replace Na₂-fumarate with glycerol 2.3 g/L; anaerobic

For <u>DSM 27333</u>: Replace Na_2 -fumarate with 2.30 g/l glycerol added after autoclaving from a sterile anoxic solution prepared under 100% N_2 gas atmosphere. Adjust pH of complete medium to 7.5.

For DSM 29728: Adjust pH of complete medium to 6.8.

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For <u>DSM 103119</u>: Replace Na_2 -fumarate with 3.2 g/l sucrose added after autoclaving from a sterile anoxic solution prepared under 100% N_2 gas atmosphere. Adjust pH of complete medium to 7.5.

Trace element solution SL-10 (from medium 320)

HCI (25%)	10.00	ml
FeCl ₂ x 4 H ₂ O	1.50	g
ZnCl ₂	70.00	mg
$MnCl_2 \times 4 H_2O$	100.00	mg
H_3BO_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
$Na_2MoO_4 \times 2 H_2O$	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
$Na_2SeO_3 \times 5 H_2O$	3.00	mg
$Na_2WO_4 \times 2 H_2O$	4.00	mg
Distilled water	1000.00	ml

Wolin's vitamin solution (10x) (from medium 120)

Biotin	20.00	mg
Folic acid	20.00	mg
Pyridoxine hydrochloride	100.00	mg
Thiamine HCI	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