829. DESULFUROBACTERIUM MEDIUM

Sea Salts (SIGMA)	30.00	g
NH_4Cl	1.00	g
KH_2PO_4	0.35	g
MES [2-(N-morpholino) ethane sulfonic acid]	1.95	g
Trace element solution SL-10 (see medium 320)	1.00	ml
Selenite-tungstate solution (see medium 385)	1.00	ml
Na-Resazurin solution (0.1% w/v)	0.50	ml
Sulfur, powdered	10.00	g
Na_2CO_3	0.50	g
Vitamin solution (see medium 503)	1.00	ml
Growth-stimulating factors (see below)	1.00	ml
Na-dithionite ($Na_2S_2O_4$)	20.00	mg
Distilled water	1000.00	ml

Dissolve ingredients (except sulfur, carbonate, vitamins, growth-stimulating factors and dithionite), boil medium for 1 min, then cool to room temperature under 80% H_2 and 20% CO_2 gas atmosphere. Adjust pH to 6.0 and dispense under 80% H_2 and 20% CO_2 gas atmosphere into anoxic Hungate-type tubes or serum vials which contain already the appropriate amount of sulfur. Autoclave at a temperature of **110°C** for 20 min! Add vitamins, growth-stimulating factors and dithionite 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. Vitamins and dithionite are sterilized by filtration. Adjust pH of the complete medium to 6.0, if necessary. After inoculation use 1 bar overpressure of sterile 80% H_2 and 20% CO_2 gas mixture.

Growth-stimulating factors:

Isobutyric acid	5.00	g
Valeric acid	5.00	g
2-Methylbutyric acid	5.00	g
3-Methylbutyric acid	5.00	g
Caproic acid	2.00	g
Succinic acid	6.00	g
Distilled water	1000.00	ml

Dissolve ingredients, adjust pH to 9.0 with NaOH, then autoclave under 100% N_2 gas.

For DSM 16661 and DSM 21157 omit growth-stimulating factors and pressurize vials after inoculation to 2 bar with sterile 80% H_2 and 20% CO_2 gas mixture.