612: DESULFOSARCINA CETONICA MEDIUM

Solution A	962.00	ml
Solution B	20.00	ml
Solution C	10.00	ml
Solution D	10.00	ml

Solution A is sparged with 80% N_2 and 20% CO_2 gas mixture to reach a pH below 6 (at least 30 min), then distributed under the same gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclaved. Solution B is autoclaved separately under 80% N_2 and 20% CO_2 gas atmosphere. Solutions C and D are autoclaved under 100% N_2 gas atmosphere. To complete the medium appropriate amounts of solutions B to D are added to the sterile solution A in the sequence as indicated. Final pH of the medium should be 7.2 – 7.4.

Solution A

KH_2PO_4	0.70	g
NH_4Cl	0.30	g
Na_2SO_4	2.80	g
$MgCl_2$ x 6 H_2O	1.70	g
$CaCl_2$ x 2 H_2O	0.05	g
NaCl	10.00	g
Trace element solution SL-10	1.00	ml
Sodium resazurin (0.1% w/v)	0.50	ml
Distilled water	960.00	ml

Solution B

Na_2CO_3	1.00	g
Distilled water	20.00	ml

Solution C

Na-butyrate	1.20	g
Distilled water	10.00	ml

Solution D

Na_2S x 9 H_2O	0.30	g
Distilled water	10.00	ml

Trace element solution SL-10 (from medium 320)

HCl (25%)	10.00	ml
$FeCl_2 \times 4\ H_2O$	1.50	g
$ZnCl_2$	70.00	mg
$MnCl_2 \times 4\ H_2O$	100.00	mg
H_3BO_3	6.00	mg
$CoCl_2 \times 6\ H_2O$	190.00	mg
$CuCl_2 \times 2\ H_2O$	2.00	mg
$NiCl_2 \times 6\ H_2O$	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.