895: ANAEROBRANCA GOTTSCHALKII MEDIUM

$(NH_4)_2SO_4$	1.00	g
NH_4Cl	0.40	g
$Na_2S_2O_3$ x 5 H_2O	0.10	g
K_2HPO_4	0.50	g
$MgSO_4$ x 7 H_2O	0.10	g
$CaCl_2$ x 2 H_2O	0.05	g
NaCl	10.00	g
$FeSO_4$ x 7 H_2O (0.1% w/v in 0.1 N H_2SO_4)	2.00	ml
Modified Wolin's mineral solution	10.00	ml
Yeast extract (BD Bacto)	0.25	g
Tryptone (BD Bacto)	0.25	g
Sodium resazurin (0.1% w/v)	0.50	ml
L-Cysteine HCl x H_2O	0.50	g
Na_2CO_3	3.00	g
Starch (soluble)	2.00	g
Wolin's vitamin solution (10x)	1.00	ml
Distilled water	1000.00	ml

Dissolve all ingredients except cysteine, carbonate, starch and vitamins. Sparge medium with 100% N_2 gas for 30 - 45 min to make it anoxic, add cysteine and adjust pH to 7.5, then dispense medium under 100% N_2 gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. Add starch and vitamins 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 should be sterilized by filtration. Adjust pH of complete medium to 9.3 - 9.5, if necessary.

For DSM 14826: Omit starch from the medium, increase the concentration of Tryptone to 2.00 g/l and supplement medium with 4.00 g/l $Na_2S_2O_3$ x 5 H_2O added to the medium from a sterile anoxic stock solution prepared under 100% N_2 gas.

For DSM 14828: Omit starch, add 5.00 g/l D-glucose and increase the concentration of Tryptone to 2.00 g/l. Adjust pH of complete medium to 9.0.

Modified Wolin's mineral solution (from medium 141)

Nitrilotriacetic acid	1.50	g
$MgSO_4$ x 7 H_2O	3.00	g
$MnSO_4$ x H_2O	0.50	g
NaCl	1.00	g
$FeSO_4$ x 7 H_2O	0.10	g
$CoSO_4$ x 7 H_2O	0.18	g
$CaCl_2$ x 2 H_2O	0.10	g
$ZnSO_4$ x 7 H_2O	0.18	g

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$CuSO_4$ x 5 H_2O	0.01	g
$AlK(SO_4)_2$ x 12 H_2O	0.02	g
H_3BO_3	0.01	g
Na_2MoO_4 x 2 H_2O	0.01	g
$NiCl_2$ x 6 H_2O	0.03	g
Na_2SeO_3 x 5 H_2O	0.30	mg
Na_2WO_4 x 2 H_2O	0.40	mg
Distilled water	1000.00	ml

First dissolve nitrilotriacetic acid and adjust pH to 6.5 with KOH, then add minerals. Adjust final to pH 7.0 with KOH.

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

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