104b: PY + X MEDIUM

This recipe contains strain-specific modifications for *Ruminiclostridium herbifermentans* DSM 109966 *

Final pH: * 7.4
Final volume: 1000 ml

Trypticase peptone (BD BBL)	5.00	g
Meat peptone (pepsin-digested)	5.00	g
Yeast extract	10.00	g
Salt solution	40.00	ml
Sodium resazurin (0.1% w/v)	0.50	ml
L-Cysteine HCl x H_2O	0.50	g
~~D-Glucose~~	~~5.00~~	~~g~~
Cellobiose	5.00	g
Distilled water	960.00	ml

Dissolve ingredients (except cysteine and glucose) and sparge medium with 100% N_2 gas for 30 - 45 min to make it anoxic. Add cysteine and adjust pH to 7.0, then dispense medium under 100% N_2 gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. After autoclaving add glucose or any other substrate from sterile anoxic stock solutions prepared under 100% N_2 gas atmosphere. Adjust pH of complete medium to 6.8 - 7.0, if necessary.

* Replace glucose with 5.0 g/l cellobiose added to the autoclaved medium from an anoxic stock solution sterilized by filtration. Adjust pH to 7.4 using a sterile anoxic stock solution of Na_2CO_3 (5% w/v) prepared under 80% N_2 and 20% CO_2 gas atmosphere

Salt solution (from medium 104)

$CaCl_2$ x 2 H_2O	0.25	g
$MgSO_4$ x 7 H_2O	0.50	g
K_2HPO_4	1.00	g
KH_2PO_4	1.00	g
$NaHCO_3$	10.00	g
NaCl	2.00	g
Distilled water	1000.00	ml