339a. WILKINS-CHALGREN ANAEROBE BROTH (N₂/CO₂)

Dissolve 33.0 g/l dehydrated Wilkins-Chalgren medium (Oxoid CM0643) in distilled water and add 0.5 ml/l Na-resazurin solution (0.1% w/v), bring to the boil and cool to room temperature while sparging with 80% N₂ and 20% CO₂ gas mixture. Add L-cysteine-HCL x H₂O (0.3 g/l), dispense the medium into anoxic Hungate-type tubes or serum vials and autoclave.

After autoclaving supplement medium with 5.0 g/l D-glucose added from a sterile anoxic stock solution sterilized by filtration. Adjust pH of medium to 6.8 with a sterile anoxic stock solution of Na₂CO₃ (5% w/v) prepared under 80% N₂ and 20% CO₂ gas atmosphere.

Note: In case resazurin is not colourless addition of 10 - 20 mg sodium dithionite per liter (e.g. from 5% (w/v) solution, freshly prepared under N₂ and filter-sterilized) can be used to completely reduce the medium.

For DSM 14428 replace D-glucose with 5.0 g/l cellobiose prepared under 100% N₂ gas atmosphere and sterilized by filtration.

For DSM 14924 and DSM 22607 adjust pH of medium after autoclaving to 7.4 using a sterile anoxic stock solution of Na₂CO₃ (5% w/v) prepared under 80% N₂ and 20% CO₂ gas atmosphere.

For DSM 15176 and DSM 15567 omit D-glucose.

For DSM 15248 adjust pH of medium to 6.0.

For DSM 28816 replace D-glucose with 3.2 g/l Na₂-fumarate prepared under 100% N₂ gas and sterilized by filtration. Adjust pH of complete medium to 7.2.