



Main sol. 700

Na ₂ CO ₃	10.00	g
NaHCO ₃	15.00	g
NaCl	10.00	g
K ₂ HPO ₄	0.20	g
NH ₄ Cl	1.00	g
KCl	0.20	g
Na ₂ S x 9 H ₂ O	1.00	g
Yeast extract	0.50	g
Sucrose	5.00	g
Wolin's vitamin solution	10.00	ml
Modified Wolin's mineral solution II	1.00	ml
Distilled water	1000.00	ml

1. Prepare the medium under N₂, omitting Na₂CO₃, Na HCO₃, Na₂S, sucrose and vitamin solution.
2. Boil the medium and cool under nitrogen, add the Na₂CO₃ and NaHCO₃ and adjust the pH to 9.7 with 6N NaOH (about 15 ml). Sterilize by autoclaving and add the Na₂S (neutralized), vitamins and sucrose from sterile stock solutions.