

**1683. CM1A Halorubrum Medium**

30% Salt water solution (see below)	829.0	ml
23% MGM (see below)	5.0	ml
Sodium pyruvate	5.5	g
Distilled water	155.0	ml

Adjust pH to 7.5 with 1 M HCl or 1 M NaOH  
Autoclave at 121 °C for 20 min.

Before inoculation add from sterile stock solutions:

NH <sub>4</sub> Cl (1M)	5.00	ml
K <sub>2</sub> HPO <sub>4</sub> (0.5M)	2.00	ml
Trace metal SL-10 (see below)	1.00	ml
Vitamin solution Vit 10 (see below)	3.00	ml

*30% Salt Water (1L):*

NaCl	240.00	g
MgCl <sub>2</sub> x 6H <sub>2</sub> O	30.00	g
MgSO <sub>4</sub> x 7H <sub>2</sub> O	35.00	g
CaCl <sub>2</sub> x H <sub>2</sub> O	0.74	g
Distilled water	to 1000.00	ml

Dissolve salts completely then add CaCl<sub>2</sub> x H<sub>2</sub>O.  
Adjust pH to 7.5 with Tris-HCl at pH=7.5

*23% MGM (100mL)*

30% Salt Water	76.60	ml
Pepton (VWR J636-500g)	0.50	g
Yeast extract (Fisher BioReagents BP1422-500)	0.10	g
Distilled water	23.40	ml

*Don't use Difco Bacto-peptone, it was reported in 1988 to contain bile salts that lyse halobacteria, this was still the case in 2001*

*Adjust pH to 7.5*

*Trace element solution SL-10:*

HCl (25%; 7.7 M)	10.00	ml
FeCl <sub>2</sub> x 4 H <sub>2</sub> O	1.50	g
ZnCl <sub>2</sub>	70.00	mg
MnCl <sub>2</sub> x 4 H <sub>2</sub> O	100.00	mg
H <sub>3</sub> BO <sub>3</sub>	6.00	mg
CoCl <sub>2</sub> x 6 H <sub>2</sub> O	190.00	mg
CuCl <sub>2</sub> x 2 H <sub>2</sub> O	2.00	mg
NiCl <sub>2</sub> x 6 H <sub>2</sub> O	24.00	mg
Na <sub>2</sub> MoO <sub>4</sub> x 2 H <sub>2</sub> O	36.00	mg
Distilled water	990.00	ml

First dissolve FeCl<sub>2</sub> in the HCl, then dilute in water, add and dissolve the other salts. Finally make up to 1000.0 ml.

*Vitamin solution 1, filter sterilized (per liter) = Vit 1:*

p-Aminobenzoic acid	40.0	mg
Biotin	10.0	mg
Nicotinic acid	100.0	mg
Calcium D-(+)-pantothenate	50.0	mg
Pyridoxamine hydrochloride	150.0	mg
Thiamine hydrochloride	100.0	mg
Cyanocobalamin (B12)	50.0	mg

*Vitamin solution 2, filter sterilized (per liter) = Vit 2:*

DL-6,8-thioctic acid	10.0	mg
Riboflavin	10.0	mg
Folic acid	4.0	mg

*Vit 10:*

mix 0.25 ml Vit 1 and 0.75 ml Vit2