High salinity medium
(for 150; make stock at 300ppt)

(Park 2012 *Extremophiles*)

Medium V (g/L)	**150ppt**	**300ppt**
NaCl	137.6	275.2
KCl	3.8	7.6
MgCl2•6H2O	13.45	26.9
MgSO4•7H2O	1.65	3.3
CaCl2•2H2O	0.65	1.3

Fill with RO water/dH2O to 1L line, **not** add 1L (~900mL instead)

Uses:

100G and 150G (G = autoclaved barley grain)
For Trimyema (microaerophile) -- 12mL 150ppt + 2 barley grains

ATCC medium: 802 Sonneborn's *Paramecium* medium

Rye grass Cerophyll:
Cerophyll*.....2.5 g
Distilled water.....1.0 L

Add cerophyll to distilled water and boil for 5 minutes. Add 100 ml distilled water to compensate for evaporation. Filter through Whatman #1 filter paper and add 0.5 g Na2HPO4. Autoclave for 15 minutes at 121C.

SW + 1%LB (SL)

SW + 3%LB (24mL LB in 800mL autoclaved SW)

Medium TZ

(based on Mesbah et al. 2007 IJSEM)

TZ160 (soda lake medium; quite caustic, wear gloves!) [based on Mesbah et al. 2007 IJSEM]

in g/L; fill to 1L (not “add 1L!”) -- make 1L mark on flask first

KH2PO4	0.2
MgCl2	0.1
KCl	0.2
NH4Cl	0.5
NaCl	100
Na2CO3	68
NaHCO3	38

Comes out to ~160-170ppt, pH 9-10, dilute to 20 ppt (measuring with refractometer)
Pour 10mL into 50mL tissue culture flask on its side with a handful (~5-8ish) autoclaved wheat grains as substrate for bacteria

Resazurin

Stock: 0.2% w/v in dH2O
final concentration to be 0.0002% w/v, stock is thus 1000x
filter sterilised and aliquoted into eppies, kept in fridge, in foil (dark)

https://link.springer.com/article/10.1023/A:1023674315047

F/2 Medium

Directions

For I L Total

1. To approximately 950 mL of non-pasteurized seawater (30-35ppt), add each of the components in the order specified (except vitamins) while stirring continuously.

2. Bring total volume to 1 L with non-pasturized seawater.

*For 1.5% agar medium add 15 g of agar into the flask; do not mix.

3. Cover and autoclave medium.

4. When cooled add sterile vitamins.

*For agar medium add vitamins, mix, and dispense before agar solidifies.

5. Store at refrigerator temperature.

#	Component	Amount	Stock Solution Concentration	Final Concentration
1	$NaNO_3$ (Fisher BP360-500)	1 mL	7.5 g/100 mL dH20	880 µM
2	$NaH_2PO_4 \cdot H_2O$(MCIB 742)	1 mL	0.5 g/100 mL dH20	36 µM
3	$Na_2SiO_3 \cdot 9H_2O$ (Sigma 307815)	1 mL	3 g/100 mL dH20	106 µM
4	Trace Metals Solution	1 mL/L		
5	Vitamin B_{12}	1 mL/L		
6	Biotin Vitamin Solution	1 mL/L		
7	Thiamine Vitamin Solution	1 mL/L		

Trace Metals Solution

Directions

For 1 L Total

1. Begin stirring and heating approximately 950 mL of dH_2O.

2. When the stirring water approaches the boiling point, add the following components in the order listed.

3. Bring the volume to 1 L with dH_2O.

4. Store at refrigerator temperature.

#	Component	Amount	Stock Solution Concentration	Final Concentration
1	$ZnSO_4 \cdot 7H_2O$ (Sigma Z 0251)	23 mg/L		0.08 µM
2	$MnSO_4 \cdot H_2O$ (Sigma M8179)	152 mg/L		0.9 µM
3	$Na_2MoO_4 \cdot 2H_2O$ (J.T. Baker 3764)	7.3 mg/L		0.03 µM
4	$CoSO_4 \cdot 7H_2O$ (Baker 1696)	14 mg/L		0.05 µM
5	$CuCl_2 \cdot 2H_2O$ (Fisher C-455)	6.8 mg/L		0.04 µM
6	$Fe(NH_4)_2(SO_4)_2 \cdot 6H_2O$ (Sigma F-1513)	4.6 g/L		11.7 µM
7	$Na_2EDTA \cdot 2H_2O$ (Sigma ED255)	4.4 g/L		11.7 µM

WC Medium

Directions

For 1 L Total:

1. To approximately 900 mL of dd-H_2O, add the following components in the orer listed (not including the vitamins) while stirring continuously.

2. Adjust the pH to 7.8.

3. Transfer the contents of the beaker to a 1-Liter graduated cylinder and bring the total volume to 1.0 Liter with dd-H_2O.

4. Add the sterile vitamin components and mix well.

5. **Do not autoclave medium! Filter sterilize only.**

6. Store at refrigerator temperature.

#	Component	Amount	Stock Solution Concentration	Final Concentration
1	$NaNO_3$ (Fisher BP360-500)	1 mL/L	85.1 g/L	1 mM
2	$CaCl_2 \cdot 2H_2O$ (Sigma C-3881)	1 mL/L	36.76 g/L	0.25 mM
3	$MgSO_4 \cdot 7H_2O$ (Sigma 230391)	1 mL/L	36.97 g/L	0.15 mM
4	$NaHCO_3$ (Fisher S 233)	1 mL/L	12.6 g/L	0.15 mM
5	$Na_2SiO_3 \cdot 9H_2O$ (Sigma 307815)	1 mL/L	28.42 g/L	0.1 mM
6	K_2HPO_4 (Sigma P 3786)	1 mL/L	8.71 g/L	0.05 mM
7	H_3BO_3 (Baker 0084)	1 mL/L	24 g/L	0.39 mM
8	WC Trace Elements Solution	1 mL/L		
9	Vitamin B_{12}	1 mL/L		
10	Thiamine Vitamin Solution	1 mL/L		
11	Biotin Vitamin Solution	1 mL/L		

From Lise Forget in Franz Lang's group: (sent 29.09.14)

WCG and WCL medium, for many protists

WCL medium seems to give better growth than WCG medium, in most cases.

To one liter of distilled and autoclaved water add:

		WCG	WCL
K2HPO4*	(8.71 g/l)	0.25 ml	----
CaCl2x2H2O	(36.76 g/l)	0.1 ml	1.0 ml
MgSO4x7H2O	(36.97 g/l)	0.25 ml	1.0 ml
NaNO3*	(75.0 g/l)	0.29 ml	1.0 ml
NaHCO3*	(12.6 g/l)	0.05 ml	1.0 ml
H3BO3*	(6.0 g/l)	0.25 ml	0.5 ml
KCL*	(7.45 g/l)	----	1.0 ml
NaH2PO4xH2O	(5.0 g/l)	----	1.0 ml
NH4CL*	(2.65 g/l)	----	1.0 ml
Na2EDTA*	(100mM Stock)	0.05 ml	----
Fe/EDTA/trace metals from F/2 stock*		0.25 ml	1.0 ml
F/2 vitamins*		0.5 ml	1.0 ml

(see on F/2 page)

In order to obtain high cell yields, buffering with HEPES is strongly recommended,
HEPES buffer: stock of 1M, adjusted to pH 7.5, end concentration in medium should be about 10-20 mM.

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Misc notes

FeCl3 (ferric citrate, oxidised state) stock: ~400mM 10^4x: 3g/50mL dH2O, filter sterilised to 1000x in sterile water and then diluted to 100x working solution (appears to be shelf-stable, the higher concentration stocks precipitate), add 200uL / 12mL of culture