L1 Medium

Marine dinoflagellates

Stocks

		per litre
(1)	$NaNO_3$	75g
(2)	$NaH_2PO_4.2H_2O$	5.65g

(3) Trace elements: There are 11 chemicals. Prepare Primary stock solutions first. Add first two chemicals separately in **500ml/dH₂O**, allowing each to completely dissolve before adding the primary stocks in order:

	Chemical	**Primary stock solutions** **10ml dH_2O**	**Amount/Volume for <u>500ml</u> working stock solution**
1	$Na_2EDTA{\cdot}2H_2O$	-	2.18g
2	$FeCl_3{\cdot}6H_2O$	-	1.575g
3	$CuSO_4{\cdot}5H_2O$	0.0245g	0.125ml
4	$Na_2MoO_4{\cdot}2H_2O$	0.199g	1.5ml
5	$ZnSO_4{\cdot}7H_2O$	0.22g	0.5ml
6	$CoCl_2{\cdot}6H_2O$	0.1g	0.5ml
7	$MnCl_2{\cdot}4H_2O$	1.8g	0.5ml
8	H_2SeO_3	0.026g	0.5ml
9	$NiSO_4{\cdot}6H_2O$	0.027g	0.5ml
10	Na_3VO_4	0.0184g	0.5ml
11	K_2CrO_4	0.0194g	0.5ml

(4) Vitamin mix:

	per litre
Cyanocobalamin (Vitamin B_{12})	0.0005 g
Biotin	0.0005 g
Thiamine HCl (Vitamin B_1)	0.1g

Vitamin mix continued:

For ease of measuring first prepare primary stock solutions.
Into 950 ml of deionised water, dissolve the thiamine HCl and add 1 ml of the primary stocks. Bring volume to 1 litre with deionised water. Filter sterilise and store frozen.

Component	Primary Stock solution	Quantity	Molar Concentration in final medium
Thiamine HCl (B_1)	-	100mg	2.96×10^{-7}
Biotin	0.5g L^{-1} dH_2O	1ml	2.05×10^{-9}
Cyanocobalamin (B_{12})	0.5g L^{-1} dH_2O	1ml	3.69×10^{-10}

Medium	**per litre**
Stock solution 1	1.0 ml
Stock solution 2	1.0 ml
Stock solution 3 (Trace elements)	1.0 ml
Stock solution 4 (Vitamin mix)	1.0 ml

Make up to 1 litre with filtered natural seawater. Adjust pH to **8.0** with 1M NaOH or 1 M HCl prior to autoclaving. For agar, add 15 g per litre Bacteriological Agar.
Autoclave at 15 psi for 15 minutes.

Reference

"*Stichochrysis immobilis* is a diatom, not a chrysophyte" Guillard and Hargraves (*Phycologia* 32.3(1993):234-236) – adapted for CCAP

Reviewed: 17th January 2025