Jun 12, 2019

K/2 Ian / K-ET

Ian Probert[1]

[1]CNRS-Sorbonne Université, Station Biologique, Place G. Tessier 29680 Roscoff FRANCE

Working dx.doi.org/10.17504/protocols.io.s53eg8n

Roscoff Culture Collection

Daniel Vaulot
Station Biologique, Roscoff, France

ABSTRACT

Modified from K medium by Ian Probert

Reference
Keller, M.D., Selvin, R.C., Claus, W. & Guillard, R.R.L. 1987. Media for the culture of oceanic ultraphytoplankton. J. Phycol. 23:633–8.

1 ▪ To 994 ml of seawater (pH 8.2, adjusted with NaOH) add:

Quantity	Compound	Stock solution (sterile)	Final conc. in K medium
0.5ml	$NaNO_3$	48.9542g/litre H_2O	288µM
0.5ml	NH_4Cl *	0.535g/litre H_2O	5µM
0.5ml	KH_2PO_4	4.8992g/litre H_2O	18µM
0.5ml	FeEDTA solution	(see recipe below)	(see below)
0.5ml	Trace metal solution	(see recipe below)	(see below)
1.0ml	f/2 vitamin solution	(see recipe below)	(see below)

* optional

FeEDTA solution

2 ▪ To 950ml distilled H_2O add:

Quantity	Compound	Stock solution	Final conc. in K medium
4.3g	(Na)FeEDTA	-	5.85µM

▪ Make up to 1 litre with milliQ H_2O, sterilize (filter 0.22µm) and store in fridge.

Trace metal solution

3 ▪ To 950ml distilled H_2O add:

Quantity	Compound	Stock solution	Final conc. in K medium
37.22g	$Na_2EDTA.2H_2O$	-	50µM
1.0ml	$CuSO_4.5H_2O$	2.497g/litre H_2O	0.005µM
1.0ml	$Na_2MoO_4.2H_2O$	7.2585g/litre H_2O	0.015µM

1.0ml	$ZnSO_4.7H_2O$	23.0g/litre H_2O	0.004µM
1.0ml	$CoSO4.7H_2O$	14.055g/litre H_2O	0.025µM
1.0ml	$MnCl_2.4H_2O$	178.11g/litre H_2O	0.45µM
1.0ml	H_2SeO_3	1.29g/litre H_2O	0.005µM
1.0ml	NiCl2.6H20	1.49 g/litre H_2O	0.00314µM

- Make up to 1 litre with milliQ H_2O, sterilize (filter 0.22µm) and store in fridge.

f/2 Vitamin solution

4 - To 950ml distilled H_2O add

Quantity	Compound	Stock solution	Final conc. in K medium
1.0ml	Vit. B_{12} (cyanocobalamin)	0.5g/litre H_2O	0.37nM
1.0ml	Biotin	5.0mg/litre H_2O	2.0nM
100.0mg	Thiamine HCl	-	0.3µM

- Make up to 1 litre with milliQ H2O, filter sterilize into plastic vials and store in freezer.

Sterilization of medium

5 - Optional: *Heat to 80°C for 2 hours and leave to cool – this should kill most organisms but should not chemically modify the medium too much*
- Filter sterilize through 0.22µm filters (e.g. Millipore Steritop units) into sterile (autoclaved) polycarbonate bottles.

For K-ET

6 - Add 10-30 ml marine soil extract (ET)