Jun 12, 2019

K and K+Si medium

DOI
dx.doi.org/10.17504/protocols.io.s34egqw

Roscoff Culture Collection[1]

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

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Daniel Vaulot
Station Biologique, Roscoff, France , Nanyang Technologica...

DOI: dx.doi.org/10.17504/protocols.io.s34egqw

Protocol Citation: Roscoff Culture Collection 2019. K and K+Si medium. **protocols.io** **https://dx.doi.org/10.17504/protocols.io.s34egqw**

Manuscript citation:

Protocol status: Working
We use this protocol and it's working

Created: August 29, 2018

Last Modified: June 12, 2019

Protocol Integer ID: 15196

Abstract

Medium to grow most phytoplankton species in small eukaryotes (Mamiellophyceae, Pelagophyceae etc...). When using for diatoms and coccolithophorids add silica (Si).

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.

Before start

Please refer to our general recommendations to grow cultures :

https://www.protocols.io/private/A48906DC1374AD6281495CB86A8F092F

1 Filter 1L of old seawater of at least two months on prefilter and filter 0,2 microns

2 Heat seawater during 20min at 100°C

3 Under hood, to seawater, add these nutriments that have been autoclaved (excepted vitamin) :

	Quantity	*Compound*	*Stock Solution*
	1.0 mL	Sodium Nitrate ($NaNO_3$)	75.0 g/L of H_2O
	1.0 mL	Ammonium Chloride (NH_4Cl)	2.68 g/L of H_2O
	1.0 mL	Na2-Glycérophosphate ($C_3H_7O_6PNa_2$, 5 à 6 H_2O)	3.06 g/L of H_2O
	1.0 mL	TRIS-Base (pH7.2)	121.1g/L of H_2O
	1.0 mL	K Trace Metal Solution	(see recipe below)
	0.1 mL	f/2 Vitamin Solution	(see recipe below)
	For K+Si	**Add**	
	1.0 mL	Sodium Metasilicate Nonahydrate ($Na_2SiO_3.9H_2O$)	30 g/L of H_2O

4 Filter the medium on 0.2 µm.

5 **K Trace Metal Solution**

- To 900 mL of H_2O add :

	Quantity	*Compound*	*Stock Solution*
	41,6g	EDTA Disodium Salt Dihydrate ($Na_2EDTA.2H_2O$)	
	3,15g	Hexahydrated ferric chloride ($FeCl_3.6H_2O$)	1,5 cc (liquide)
	1.0 ml	Sodium Molybdate Dihydrate ($Na_2MoO_4.2H_2O$)	6,3 g/L of H_2O
	1.0 ml	Zinc Sulfate Heptahydrate ($ZnSO_4.7H_2O$)	22.0g/L of H_2O
	1.0 ml	Cobalt Chloride Hexahydrate ($CoCl_2.6H_2O$)	10.0g/L of H_2O
	1.0 ml	Manganese (II) chloride, tetrahydrate ($MnCl_2.4H_2O$)	180.0g/L of H_2O
	1.0 ml	Copper(II) sulfate pentahydrate (Cu $SO_4.5H_2O$)	4.9g/L of H_2O
	1.0 ml	Selenous acid (H_2SeO_3)	1,29g/L of H_2O

- Make final volume up to 1.0L using H2O. Heat to dissolve

6 **f/2 Vitamin solution**

- In 100mL of distilled water, dissolve 0,05g of biotin (vit. H) and 0,05g of cyanocobalamin (vit. B_{12}) = solution A
- In 0,5mL of solution A, add 0,05g of thiamine HCl (vit. B_1) and complete final volume to 50mL of distilled water
- Filter sterilize on Millipore 0,22µm
- Store in refrigerator or freezer