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🌐 PCC559 medium

Roscoff Culture Collection¹

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

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ABSTRACT

Pasteur Collection medium for cyanobacteria (*Prochlorococcus*, *Synechococcus*).

Not used by the RCC.

BEFORE START INSTRUCTIONS

Please refer to our general recommendations to grow cultures :

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

Medium composition

- 1 ■ Under hood, add these nutriments autoclaved before (excepted vitamin):

<i>Quantity</i>	<i>Compound</i>
1 L	Turks Island Salts 1X
4 mL	Ferric chloride hexahydrate/EDTA solution
1mL	Trace metal for Prochlorococcus medium
1 mL	Na-PO4 solution (50 mM, pH 7.5)
4 mL	Ammonium sulfate solution (100 mM)
2 mL	Sodium hydrogenocarbonate solution (1 M)
1 mL	Vitamin B12 (Cyanocobalamin)

- Filter the medium on 0,2microns

Turks Island Salts 1X

- 2 ■ Dissolve these salts in the volume of water indicated

<i>Quantity</i>	<i>Compound</i>	<i>Volume of dissolution</i>
28 g	Sodium chloride (NaCl)	450 mL
670 mg	Potassium chloride (KCl)	50 mL
5,5 g	Magnesium chloride hexahydrate (MgCl ₂ ·6H ₂ O)	100 mL
6,9 g	Magnesium sulfate heptahydrate (MgSO ₄ ·7H ₂ O)	150 mL
1,45 g	Calcium chloride dihydrate (CaCl ₂ ·2H ₂ O)	100 mL

- Mix the solutions in the order indicated
- Complete final volume to 1L of distilled water
- Autoclave
- Store in refrigerator

Ferric chloride hexahydrate/EDTA solution

- 3 ■ To 10mL of HCl 0,1N, add gradually 270mg of Ferric chloride hexahydrate (FeCl₃·6H₂O)
- To 10mL of NaOH 0,1N, add gradually 372mg of Titriplex III dihydrate (EDTA-Na₂)
- Mix both solutions
- Complete final volume to 500mL of sterile water
- Store in refrigerator

Trace metal for Prochlorococcus medium

4 ■ Dissolve these salts in the volume of water indicated :

<i>Quantity</i>	<i>Compound</i>	<i>Volume of dissolution</i>
2.86 g	Boric acid (H ₃ BO ₃)	150 mL
1.81 g	Manganese chloride tetrahydrate (MnCl ₂ ·4H ₂ O)	150 mL
0.222 g	Zinc sulfate heptahydrate (ZnSO ₄ ·7H ₂ O)	150 mL
0.39 g	Sodium molybdate dihydrate (Na ₂ MoO ₄ ·2H ₂ O)	300 mL
0.049 g	Cobalt(II) nitrate hexahydrate (Co(NO ₃) ₂ ·6H ₂ O)	150 mL

- Mix the solutions in the order indicated
- Complete final volume to 1L of sterile water
- Store 6 months in refrigerator

Safety information

Attention: dilute 10 times with sterile water and filter on 0.2 microns before use

Other solutions

5 **Na-PO₄ solution (50 mM, pH 7.5)**

- Prepare two solutions :
- Dissolve 3,45g of monosodium dihydrogen phosphate (NaH₂PO₄) in 50mL of water
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- Dissolve 4,45g of di-sodium hydrogenophosphate dihydrate (Na₂HPO₄·2H₂O) in 50mL of water
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- Make an equimolar mixture of this two solutions and adjust the pH at 7,5
- Store in refrigerator

Ammonium sulfate solution (100 mM)

- To 250mL of distilled water, add 3,3g of ammonium sulfate ((NH₄)₂SO₄)
- Autoclave the solution
- Store in refrigerator

Sodium hydrogenocarbonate solution (1 M)

- To 300mL of distilled water, add 25,2g of sodium hydrogenocarbonate (NaHCO₃)
- Autoclave the solution
- Store in refrigerator

Vitamin B12 (Cyanocobalamin)

- To 20mL of distilled water, add 20mg of vitamin B12
- Filter on 0,2 microns (=stock solution at 1mg/mL)

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- Store in freezer
 - The working solution (10µg/mL) is prepared by aseptical dilution of the vitamin B12 stock solution with sterile water