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Provasoli Enriched Seawater (PES) medium solution

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Protocol status: Working

We use this protocol and it's working

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Abstract

This protocol outlines the steps required to prepare Provasoli Enriched Seawater (PES) medium in accordance with the method by Provasoli (1966).

CITATION

L. Provasoli (1966). Media and prospects for the cultivation of marine algae. Proceedings of the US-Japan Conference.

LINK

<https://cir.nii.ac.jp/crid/1572824499547489536>















Protocol materials

- ⊗ MilliQ water In [6 steps](#)
- ⊗ Cobalt(II) sulfate heptahydrate **Merck MilliporeSigma (Sigma-Aldrich)** Step 1.5
- ⊗ Titriplex® III solution **Merck MilliporeSigma (Sigma-Aldrich)** In [2 steps](#)
- ⊗ Zinc sulfate heptahydrate **Merck MilliporeSigma (Sigma-Aldrich) Catalog #204986** Step 1.4
- ⊗ Iron(III) chloride hexahydrate **Merck MilliporeSigma (Sigma-Aldrich) Catalog #44944** Step 1.6
- ⊗ Ammonium iron(II) sulfate hexahydrate **Bio Basic Inc. Catalog #AB0065.SIZE.500g** Step 2.2
- ⊗ Biotin **P212121** Step 4.3
- ⊗ Sodium nitrate **Merck MilliporeSigma (Sigma-Aldrich) Catalog #S5506** Step 3.4
- ⊗ Vitamin B12 **Fisher Scientific Catalog #68-19-9** Step 4.1
- ⊗ Sodium glycerophosphate hydrate **Merck MilliporeSigma (Sigma-Aldrich)** Step 3.5
- ⊗ Boric acid **Fisher Scientific Catalog #BP1681** Step 1.2
- ⊗ Manganese sulfate, monohydrate **Bio Basic Inc. Catalog #MB0334.SIZE.500g** Step 1.3
- ⊗ HEPES Sodium salt **Merck MilliporeSigma (Sigma-Aldrich) Catalog #H7006** Step 3.1
- ⊗ Thiamine HCl **P212121** Step 4.2
- ⊗ Potassium iodide Step 3.6







Before start

It is important to follow the order of chemicals added as shown in the protocol.

P II solution

- 1  5000 mL  MilliQ water **Sigma Aldrich**
- 1.1 Add  5 g  Titriplex® III solution **Sigma Aldrich**
- 1.2 Add  5.7 g  Boric acid **Sigma Aldrich Catalog #BP1681**
- 1.3 Add  0.62 g
 Manganese sulfate, monohydrate **Sigma Aldrich Catalog #MB0334.SIZE.500g**
- 1.4 Add  0.11 g  Zinc sulfate heptahydrate **Sigma Aldrich Catalog #204986**
- 1.5 Add  0.024 g  Cobalt(II) sulfate heptahydrate **Sigma Aldrich**
- 1.6 Add  0.245 g  Iron(III) chloride hexahydrate **Sigma Aldrich Catalog #44944**

Fe-solution

- 2  5000 mL  MilliQ water **Sigma Aldrich**
- 2.1 Add  3.3 g  Titriplex® III solution **Sigma Aldrich**
- 2.2 Add  3.51 g
 Ammonium iron(II) sulfate hexahydrate **Sigma Aldrich Catalog #AB0065.SIZE.500g**

**Note**Solution equals $0.1 \text{ mg Fe}^{2+} \times \text{ml}^{-1}$ **Primary solution**










2h

- 3
- 3.1 Add (or Tris buffer)
- 3.2 Add P II-solution
- 3.3 Add Fe-solution
- 3.4 Add
- 3.5 Add
- 3.6 Add
- 3.7 Pasteurise for at


2h

Vitamins

- 4 Vitamins to be added through a sterile single injection with a front filter (0.2 μm) in the cooled off primary solution (maximum temperature of).

- 4.1  30 mL  Vitamin B12 **Sigma Aldrich Catalog #68-19-9** (10 mg per 500 ml
 MilliQ water **Sigma Aldrich**)
- 4.2  30 mL  Thiamine HCl **Sigma Aldrich** (500 mg per 500 ml
 MilliQ water **Sigma Aldrich**)
- 4.3  30 mL  Biotin **Sigma Aldrich** (5 mg per 500 ml  MilliQ water **Sigma Aldrich**)

Storage

- 5 Store PES in Duran bottles in a cold and dark setting at  10 °C .

Equipment

Duran® laboratory bottles, with caps	NAME
Glass bottle	TYPE
Duran	BRAND
Z305197-10EA	SKU
500 ml	SPECIFICATIONS

Application

- 6 Culturing medium is created by adding  200 mL PES to  10 L sterile seawater.

Citations

L. Provasoli. Media and prospects for the cultivation of marine algae
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