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 We use this protocol and it's working

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🌐 BG11 medium

Roscoff Culture Collection¹

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Roscoff Culture Collection

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ABSTRACT

Medium to grow freshwater cyanobacteria.

MATERIALS

STEP MATERIALS

🧪 BG11 medium **Merck MilliporeSigma (Sigma-Aldrich) Catalog #C3061-500ML**

BEFORE START INSTRUCTIONS

Please refer to our general recommendations to grow cultures :

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

Prepare using Sigma Aldrich stock

- 1 ■ Under hood, to 1L of sterile water, add 20 mL of Cyanobacteria BG-11 Freshwater Solution from Sigma

- Filter the medium on 0,2 microns

⊗ BG11 medium Merck MilliporeSigma (Sigma-Aldrich) Catalog #C3061-500ML

Prepare from base chemicals

2

Protocol

NAME

Recipe for standard BG-11 media

CREATED BY

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PREVIEW

2.1

- $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$ ($3.6 \text{ g} \cdot \text{L}^{-1}$)
- Citric acid ($0.6 \text{ g} \cdot \text{L}^{-1}$)
- NaNO_3 ($149.58 \text{ g} \cdot \text{L}^{-1}$)
- $\text{MgSO}_4 \cdot 7 \text{ H}_2\text{O}$ ($7.49 \text{ g} \cdot \text{L}^{-1}$)
- 0.25 M $\text{Na}_2\text{-EDTA}$, pH 8.0 ($0.56 \text{ ml} \cdot \text{L}^{-1}$)

For 100x BG11 Stock -N:

- Omit NaNO_3 .

2.2

- 1000x Na_2CO_3 : 20 mg L^{-1}
- 100x TES-buffer, pH 8.0 (1M), adjust with KOH
- 1000x $\text{K}_2\text{HPO}_4 \times 3 \text{ H}_2\text{O}$: $30 \text{ mg} \cdot \text{mL}^{-1}$
- 1000x Fe(III) ammonium citrate ($6 \text{ mg} \cdot \text{L}^{-1}$)
- 5000x $\text{CuSO}_4 \cdot 5 \text{ H}_2\text{O}$ ($395 \text{ ng} \cdot \text{mL}^{-1}$) (sterilize using a filter)

2.3

1000x concentration:

- H_3BO_3 ($2.86 \text{ g} \cdot \text{L}^{-1}$)
- $\text{MnCl}_2 \cdot 4 \text{ H}_2\text{O}$ ($1.81 \text{ g} \cdot \text{L}^{-1}$)
- $\text{ZnSO}_4 \cdot 7 \text{ H}_2\text{O}$ ($0.222 \text{ g} \cdot \text{L}^{-1}$)
- $\text{Na}_2\text{MoO}_4 \cdot 2 \text{ H}_2\text{O}$ ($0.390 \text{ g} \cdot \text{L}^{-1}$)
- $\text{Co}(\text{NO}_3)_2 \cdot 6 \text{ H}_2\text{O}$ ($0.049 \text{ g} \cdot \text{L}^{-1}$)

For BG11 lacking certain metals (e.g. for working with metal inducible promoters P_{petE} ,

P_{coaT} , P_{ziaA} etc., trace metal mix can be prepared lacking these chemicals and used

instead of standard trace metal mix.

2.4 Fill 1 L bottle with 500 mL ultra pure water. Add stock solutions as shown below.

Stock solution	Volume
100x BG11 Stock	10 mL
1000x Na ₂ CO ₃	1 mL
1000x K ₂ HPO ₄ x 3 H ₂ O	1 mL
100x TES-buffer	10 mL
1000x Trace Metal Mix	1 mL

Add ultra pure water to 1 L.

Autoclave.

After autoclaving, add 1 mL 1000x Fe(III) ammonium citrate.

Optional: After autoclaving, add 200 µL 5000x CuSO₄

2.5 Fill 1 L bottle with 500 mL ultra pure water. Add stock solutions as shown below.

Stock solution	Volume
100x BG11 Stock -N	10 mL
1000x Na ₂ CO ₃	1 mL
1000x K ₂ HPO ₄ x 3 H ₂ O	1 mL
100x TES-buffer	10 mL
1000x Trace Metal Mix	1 mL

Add ultra pure water to 1 L.

Autoclave.

After autoclaving, add 1 mL sterile 1000x Fe(III) ammonium citrate.

Optional: After autoclaving, add 200 µL sterile 5000x CuSO₄

2.6 Fill 500 mL bottle with 250 mL ultra pure water. Add stock solutions as shown below.

Stock solution	Volume
100x BG11 Stock -N	10 mL
1000x Na ₂ CO ₃	1 mL
1000x K ₂ HPO ₄ x 3 H ₂ O	1 mL
100x TES-buffer, pH = 8.0	10 mL
1000x Trace Metal Mix	1 mL

Add ultra pure water to 500 mL.

Autoclave.

After autoclaving, add 1 mL sterile 1000x Fe(III) ammonium citrate.

Optional: After autoclaving, add 200 μ L sterile 5000x CuSO₄

2.7 Prepare 1.5 % agar: Weigh 4.5 g Bacto Agar. Fill up to 300 mL. Autoclave.

Microwave agar until liquid. Let cool.

2.8 In a 50 mL Falcon, add 1 vol 2x BG11 and 1 vol liquid 1.5 % agar. (Note: Usually, one plate requires 30-40 mL total volume.)

2.9 When mixture is hand warm, add appropriate antibiotics, if required. Quickly pour plate, avoiding air bubble