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# BG11 medium

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Roscoff Culture Collection[1]

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Roscoff Culture Collection

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

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## Abstract

Medium to grow freshwater cyanobacteria.

## Materials

STEP MATERIALS

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

## Protocol materials

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## Before start

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

Anna Behle

PREVIEW

2.1
- $CaCl_2$ $2H_2O$ (3.6 g · $L^{-1}$)
- Citric acid (0.6 g · $L^{-1}$)
- $NaNO_3$ (149.58 g · $L^{-1}$)
- $MgSO_4$ · 7 $H_2O$ (7.49 g · $L^{-1}$)
- 0.25 M $Na_2$-EDTA, pH 8.0 (0.56 ml · $L^{-1}$)

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 $K_2HPO_4$ x 3 $H_2O$: 30 mg · $mL^{-1}$
- 1000x Fe(III) ammonium citrate (6 mg · $L^{-1}$)
- 5000x $CuSO_4$ 5 $H_2O$ (395 ng · $mL^{-1}$) (sterilize using a filter)

2.3 1000x concentration:
- $H_3BO_3$ (2.86 g · $L^{-1}$)
- $MnCl_2$ · 4 $H_2O$ (1.81 g · $L^{-1}$)

- $ZnSO_4 \cdot 7\ H_2O$ (0.222 g · $L^{-1}$)
- $Na_2MoO_4 \cdot 2\ H_2O$ (0.390 g · $L^{-1}$)
- $Co(NO_3)_2 \cdot 6\ H_2O$ (0.049 g · $L^{-1}$)

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 $Na_2CO_3$ | 1 mL |
| | 1000x $K_2HPO_4$ x 3 $H_2O$ | 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 $CuSO_4$

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 $Na_2CO_3$ | 1 mL |
| | 1000x $K_2HPO_4$ x 3 $H_2O$ | 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 $CuSO_4$

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 $Na_2CO_3$ | 1 mL |
| | 1000x $K_2HPO_4$ x 3 $H_2O$ | 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 µL sterile 5000x $CuSO_4$

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 bubbl