



2019

## **BG11** medium

Roscoff Culture Collection<sup>1</sup>

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**ABSTRACT** 

Medium to grow freshwater cyanobacteria.

STEPS MATERIALS

BG11 medium

CATALOG # NAME

> C3061-500ML Sigma - Aldrich

VENDOR

## Prepare using Sigma Aldrich stock

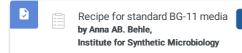
Under hood, to 1L of sterile water, add 20 mL of Cyanobacteria BG-11 Freshwater Solution from Sigma

• Filter the medium on 0,2 microns



## Prepare from base chemicals

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- 2.1 CaCl<sub>2</sub> 2H<sub>2</sub>O (3.6 g · L<sup>-1</sup>)
  - Citric acid (0.6 g ⋅ L<sup>-1</sup>)
  - NaNO<sub>3</sub> (149.58 g · L<sup>-1</sup>)
  - $MgSO_4 \cdot 7 H_2 O (7.49 g \cdot L^{-1})$
  - 0.25 M Na<sub>2</sub>-EDTA, pH 8.0 (0.56 ml · L<sup>-1</sup>)

For 100x BG11 Stock -N:

- Omit NaNO<sub>3</sub>.
- 2.2 ■ 1000x Na<sub>2</sub>CO<sub>3</sub>: 20 mg L<sup>-1</sup>

- 100x TES-buffer, pH 8.0 (1M), adjust with KOH
- $1000x K_2HPO_4 x 3 H_2O: 30 mg \cdot mL^{-1}$
- 1000x Fe(III) ammonium citrate (6 mg · L<sup>-1</sup>)
- 5000x CuSO<sub>4</sub> 5 H<sub>2</sub>O (395 ng·mL<sup>-1</sup>) (sterilize using a filter)

## 2.3 1000x concentration:

- $H_3BO_3$  (2.86 g·L<sup>-1</sup>)
- MnCl<sub>2</sub> · 4 H<sub>2</sub>O (1.81 g · L<sup>-1</sup>)
- $ZnSO_4 \cdot 7 H_2O (0.222 g \cdot L^{-1})$
- Na<sub>2</sub>MoO<sub>4</sub> · 2 H<sub>2</sub>O (0.390 g · L<sup>-1</sup>)
- $Co(NO_3)_2 \cdot 6 H_2O (0.049 g \cdot L^{-1})$

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 <sub>2</sub> CO <sub>3</sub>	1 mL
1000x K <sub>2</sub> HPO <sub>4</sub> x 3 H <sub>2</sub> 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 CuSO<sub>4</sub>

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 <sub>2</sub> CO <sub>3</sub>	1 mL
1000x K <sub>2</sub> HPO <sub>4</sub> x 3 H <sub>2</sub> 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 CuSO<sub>4</sub>

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 <sub>2</sub> CO <sub>3</sub>	1 mL
1000x K <sub>2</sub> HPO <sub>4</sub> x 3 H <sub>2</sub> 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 µL sterile 5000x CuSO⊿		
	optional. Arter dutooldving, and 200 pt oteline 0000x 00004		
2.7	Prepare 1.5 % agar: Weigh 4.5 g Bacto Agar. Fill up to 300 mL. Autoclave.		
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	Microwave agar until liquid. Let cool.		
2.8	.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		

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