Chlorophyll Extraction in Cyanobacteria Version 2

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Abstract

This protocol should be used for chlorophyll extraction in cyanobacteria. The equation for calculating the exact chlorophyll content can be found at the end of this document.

You might want to measure the optical density (OD) of you cyanobacteria culture at 750 nm. Use BG11 medium or water as the reference solution. You need the OD of your culture to normalize the cholorphyll concentration to the number of cyanobacteria.

Calculate chlorophyll content (adapted from Lichtenthaler 1978)

Chl [μ g/ml] = OD_{665nm} x 13.9 [μ g/ml] x dilution factor of culture

You can take less than 1 ml, but note the dilution factor for the calculation later on, e.g.:

1 ml sample = dilution factor of 1

500 μ l sample = dilution factor of 2

100 μ l sample = dilution factor of 10

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Protocol

Step 1.

Take 1 ml sample of your cyanobacteria culture and spin it down at 14,000 rpm for 5 min.

O DURATION

00:05:00

Step 2.

Discard 0.9 ml of the supernatant. Resuspend the pellet in the remaining 100 µl.

Step 3.

Add 0.9 ml of 100% methanol to the sample and mix thoroughly by vortexing.

Step 4.

Incubate the samples in the dark for 30 min at 4 °C in the fridge.

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Step 5.

Spin down samples again at 14,000 rpm for 5 min.

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Step 6.

Transfer supernatant into a cuvette and measure the **extinction** at **665 nm**. Use **90% methanol** as the **reference** solution.