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Chlorophyll Extraction in Cyanobacteria

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

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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 [µg/ml] = OD_{665nm} x 13.9 [µg/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 µl sample = dilution factor of 2
100 µl sample = dilution factor of 10

1 Take **1 ml sample** of your cyanobacteria culture and spin it down at **14,000 rpm** for **5 min**.
00:05:00

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

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

4 Incubate the samples in the **dark** for **30 min** at **4 °C** in the fridge.
00:30:00

5 Spin down samples again at **14,000 rpm** for **5 min**.
00:05:00

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