Monitoring fluorescence during cyanobacterial physiology/lysis experiments Version 2

Sarah Giuliani

Abstract

Monitor fluorescence of cyanobacterial cultures over time using a 96-well plate format.

Citation: Sarah Giuliani Monitoring fluorescence during cyanobacterial physiology/lysis experiments. **protocols.io** dx.doi.org/10.17504/protocols.io.f28bqhw
Published: 07 Oct 2016

Materials

- 96 well plates with lids FISHER SCIENTIFIC #087722C by Contributed by users
- P20 micropipet and filter tips by Contributed by users
- Tecan fluorescence plate reader by Contributed by users
- ASW media by Contributed by users

Protocol

Step 1.

Advance Preparation: Aliquot 180 µl seawater medium (e.g. ASW) into each well of a 96-well plate. Keep a log of your plate layout (which samples in which wells).

Plate-Setup:

	1	2	3	4	5	6	7	8	9	10	11	12
A												
B												
C												
D												
E												
F												
G												
H												

NOTES

Maureen Coleman 07 Oct 2016

This will result in a 10-fold sample dilution (20µl into 180µl) and is useful for dense cultures that would otherwise lead to "OVER" readings in the fluorometer.

Step 2.

Add 20 µl of sample into the ASW for a 10X dilution. Mix well with a pipette.

Step 3.

Read fluorescence using the Tecan Infinite Pro 200M plate reader. Set up a program using the following wavelengths: excitation 440 nm, emission 680 nm for chlorophyll a; excitation 544 nm, emission 577 nm for phycoerythrin.

Step 4.

Plot fluorescence vs. time to track the real-time progression of cell growth and/or phage infection.

NOTES

Maureen Coleman 07 Oct 2016

Keep in mind that fluorescence is not always a good proxy for cell numbers. Fluorescence can change for a variety of reasons -- physical (e.g. lysis/dissociation), physiological (e.g. nutrient limitation), etc.