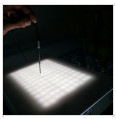

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Measuring PPFD on Algal Shaker v.2

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ABSTRACT

The illuminated orbital shaker features a glowing growing area. The illumination is not entirely homogeneous. It is brightest near the centers of the LEDs and becomes dimmer closer to the edges. The light brightness of the light in the glowing area is dependent on the LEDs, their spacing, the distance between the LEDs and the transparent platform, the material of the anti-slip mat, among other parameters. To ensure consistency and reproducibility between experiments, the brightness can be measured and the light output of the illuminator calibrated. Quantum detector is used to measure the photosynthetic photon flux density (PPFD). The measurement described in this protocol is conducted for all current settings on the LED controller and in 12 different locations across the glowing area. It is done both in air and inside an Erlenmeyer flask containing water, to best capture the actual conditions experienced by the microalgae cultures.

EXTERNAL LINK

https://app.labstep.com/sharelink/fcc4e7da-8aff-4a1f-b611-3005066f8851

GUIDELINES

The goal is to calibrate the irradiance on the surface of the microalgae shaker in air and also inside a glass flask with deionized water in it against the setting of the LEDs.

For each LED current setting, the measurement must be taken in many locations due to the inhomogeneity of the illumination pattern. The resulting value is then the average of all these measurements for each current setting.

Acknowledgement: The measurements are done using professional equipment borrowed from Giorgio Perin (Patrick Jones group) at Imperial College London.

MATERIALS TEXT

Light Meter LI-250A
Light Meter
Li-Cor Biosciences .
Part Number: LI-250A

PDF

Citation: Jakub Nedbal (03/20/2020). Measuring PPFD on Algal Shaker. https://dx.doi.org/10.17504/protocols.io.bdyxi7xn

Spherical Micro Quantum Sensor
Light Detector
Walz .
Part Number: US-SQS/L

Borosilicate Glass Narrow Neck Erlenmeyer Flasks (100 ml)
by Fisher Scientific
Catalog #: 15409103

Water refers to sterilized deionized water

SAFETY WARNINGS

The LEDs are bright at the higher current settings. The brightness of the light right at the orbital shaker platform is comparable to the Sun on a bright day. Do not stare into the light. Keep a distance between your eyes and the glowing LEDs. Use sunglasses or other protective equipment if you find your eyes becoming sensitive due to the bright light.

BEFORE STARTING

This protocol has been designed for a particular type of quantum light sensor (US-SQS/L, Walz) and light detector (LI-250A, Li-Cor Biosciences). It may need to be adjusted for different equipment.

Preparation

1 Switch off the algal shaker

Clear out any culture flasks from the algal shaker.

Move it onto a bench.

2 Work in a dark room. Turn all lights in the room off. Keep them off during the measurements.

3 Connect the Walz probe to the Li-COR Light Meter.

Set the Li-COR Light Meter calibration to measure in air (-274.3). This value is specific to the particular probe and will be different for other probes. The value in the calibration data for specifically for the utilized probe must be used.

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Measurement

4 Place the probe on a random spot on top of the illuminated shaker platform and record the value.

Hold the probe vertically upright and keep your hand as far away from the illuminator as possible to minimize effect of light scattered from the hand and sleeve.

Repeat this in twelve different random places. Find a random location by doing it with eyes closed or looking away to avoid personal bias.

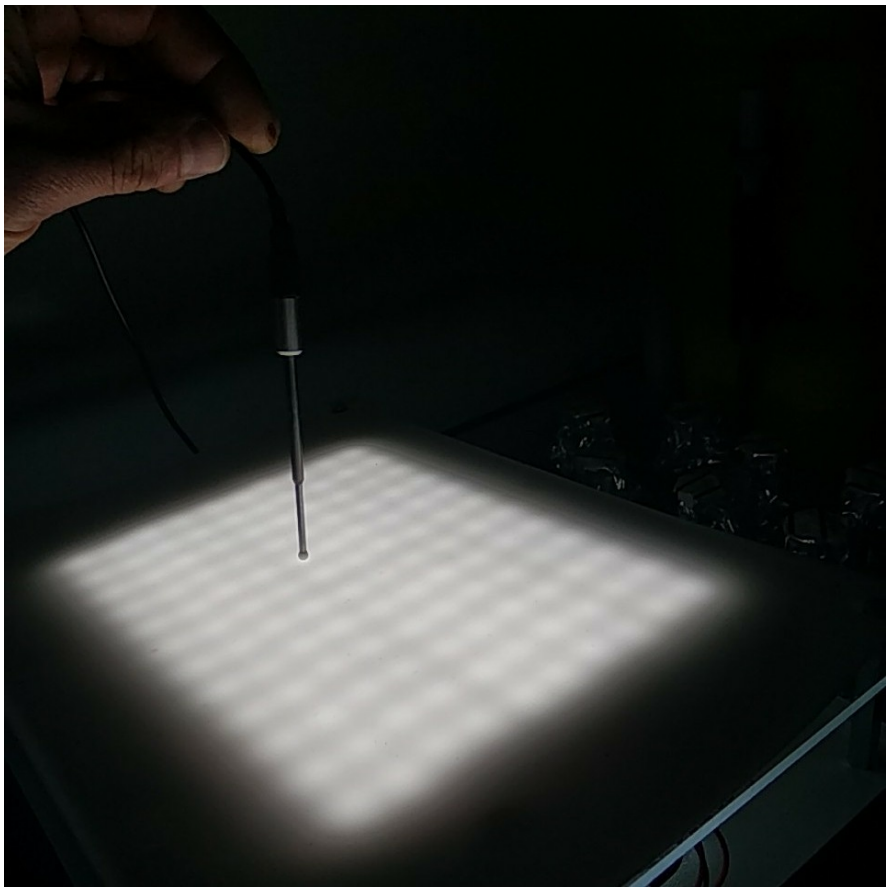

Photo showing how the proble is held perpendicularly to the surface of the illuminated orbital shaker platform with the hand as far as possible to minimize the contribution of the light scattering from the hand.

5 Repeat the above step for all eight LED current settings.

Citation: Jakub Nedbal (03/20/2020). Measuring PPFD on Algal Shaker. https://dx.doi.org/10.17504/protocols.io.bdyxi7xn

6 Record all PPFD measurements in this table:

Repeat	LED Current Setting							
	Trickle	·	1	2	3	4	5	6
1								
2								
3								
4								
5								
6								
7								
8								
9								
10								
11								
12								

Photosynthetic Photon Flux Density in Air. Table to record twelve repeats of PPFD measurements for eight LED current settings done in air on the surface of the illuminated orbital shaker platform.

7 Set the Li-COR Light Meter calibration to measure in water (-471.9). This value is specific to the particular probe and will be different for other probes. The value in the calibration data for specifically for the utilized probe must be used.

8 Place a 100 ml Erlenmeyer flask onto the illuminated shaker platform.

9 Place the probe into the flask fix the flask mouth with some cotton wool, replicating the plug normally used in the culture. Try to keep the probe vertical.

Record the value.

Move the flask to another random spot, repeating the measurement in twelve different places. Find a random location by doing it with eyes closed or looking away to avoid personal bias.

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Photo showing how the proble is held perpendicularly to the surface of the illuminated orbital shaker platform inside an Erlenmeyer flask with deionized water. Cotton wool plug in the neck of the flasks holds the probe and also reproduces the scattering from the cotton wool used to plug the flasks with the microalgae cultures.

10 Repeat the above step for all eight LED current settings.

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11 Record all PPFD measurements in this table:

Repeat	LED Current Setting							
	Trickle	·	1	2	3	4	5	6
1								
2								
3								
4								
5								
6								
7								
8								
9								
10								
11								
12								

Photosynthetic Photon Flux Density in Water. Table to record twelve repeats of PPFD measurements for eight LED current settings done in water inside an Erlenmeyer flask on the surface of the illuminated orbital shaker platform.

Analysis and Data Presentation

12 Download the MATLAB® code to analyze the PPFD measurement from GitHub:

github.com/jnedbal/PPFD

The project directory contains the following files:

File	Description
Air.pdf	Graphical representation of the PPFD results measured in air in PDF format
Air.png	Graphical representation of the PPFD results measured in air in PNG format
LICENSE	BSD 2-Clause license
PPFD.xlsx	Table with PPFD measurement results. Update the table with your own measurements
PPFD.m	MATLAB® code to analyse the measurement data in PPFD.xlsx table. Requires UnivarScater.
Water.pdf	Graphical representation of the PPFD results measured in water in PDF format
Water.png	Graphical representation of the PPFD results measured in water in PNG format

Table listing the files in the github.com/jnedbal/PPFD project.

13 Update the PPFD.xlsx file downloaded in the above step with the actual measurement values obtained from your illuminated orbital shaker.

Citation: Jakub Nedbal (03/20/2020). Measuring PPFD on Algal Shaker. https://dx.doi.org/10.17504/protocols.io.bdyxi7xn

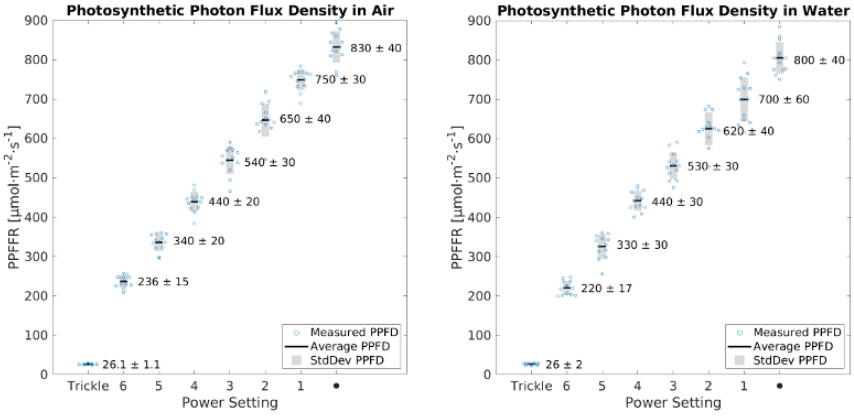
14 PPFD requires function UnivarScatter downloaded from Mathworks® File Exchange:

https://uk.mathworks.com/matlabcentral/fileexchange/54243-univarscatter

The UnivarScatter project was written by Manuel Lera Ramírez.

Place the files from the UnivarScatter project in the same folder as PPFD.m on your computer, or put it into another path, where MATLAB® can find it. You can use addpath function for that purpose. Without UnivarScatter, PPFD will throw an error.

15 Run PPFD in MATLAB®. The graphs will be produced according to the measurement values in PPFD.xlsx spreadsheet.



Example of the graphs generated by PPFD.m based on the measurement data in PPFD.xlsx.

References

16 **References**

- LI-250A Instruction Manual
- US-SQS/L Instruction Manual
- PPFD project: Code to present the results of the PPFD measurement in a graphical format.
- UnivarScatter project: MATLAB® code by Manuel Lera Ramírez used to produce univariate scatter plots.

Citation: Jakub Nedbal (03/20/2020). Measuring PPFD on Algal Shaker. https://dx.doi.org/10.17504/protocols.io.bdyxi7xn