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Microplate Reader Workflow V.2

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1 Works for me dx.doi.org/10.17504/protocols.io.8bqhsmw

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1 Refresh overnight cell culture in LB media at 37 °C

Preparation of LB Media
by NUS iGEM,
National University of Singapore
PREVIEW RUN

1.1 Weigh 25 g of Luria Broth Base powder.

Luria Broth Base (Miller's LB Broth Base)™, powder
by Thermo Fisher
Catalog #: 12795027

1.2 Add the powder into 1 L of water.

Water refers to sterilized deionized water

1.3 Autoclave entire bottle of LB media.

2 Measure OD_{600} of cell culture for desired OD value

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3 Load **200 µl** of cell samples into three wells of a 96-well plate (triplicates)

4 Induce appropriate volumes of chemical inducers (not exceeding **6 µl**) in each well

5 Include LB media as blanks

6 Load the plate (with lid) into microplate reader

Cytation 5
Automated microscope; Multi-mode plate reader

BioTek N.A.

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7 Start protocol to run continuous measurement overnight

OD_{600} (absorbance) protocol:
Wavelength: 600nm
Measurement interval time: 8min
Run time: 18h

GFP (fluorescence) protocol:
Excitation: 485 +/- 10
Emission: 528 +/- 10
Gain: Extended

RFP (fluorescence) protocol:
Excitation: 535 +/- 20
Emission: 600 +/- 20
Gain: Extended

Luminescence protocol:
Gain: 118
Integration time: 1s

OD_{600} + GFP protocol script:
Measurement interval time: 8min
Run time: 18h

OD_{600} + luminescence protocol script:
Measurement interval time: 20min
Run time: 20h

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