May 24, 2024

🌐 A large scale bacterial attraction assay: a new quantitative bacterial migration assay suitable for genetic screens

📖 PLOS One

✓ Peer-reviewed method

Thomas Quiroz Monnens[1], Alice Boulanger[1]

[1]LIPME, Université de Toulouse, INRAE, CNRS, Université Paul Sabatier, 31320, Castanet-Tolosan, France

| PLOS ONE Lab Protocols | Spotlight series |

👤 **Boulanger Alice**
université Paul sabatier - toulouse III

Create & collaborate more with a free account

Edit and publish protocols, collaborate in communities, share insights through comments, and track progress with run records.

Create free account

Collection Citation: Thomas Quiroz Monnens, Alice Boulanger 2024. A large scale bacterial attraction assay: a new quantitative bacterial migration assay suitable for genetic screens. **protocols.io**
https://dx.doi.org/10.17504/protocols.io.n2bvjn65bgk5/v1

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

Created: May 03, 2024

Last Modified: October 09, 2024

Collection Integer ID: 99639

Keywords: large scale bacterial attraction assay, new quantitative bacterial migration assay, bacterial attraction assay, genetic screens bacteria, attracted bacteria, large quantities of attracted bacteria, bacterial migration, motile bacterial cell, high numbers of attracted bacteria, rapid chemotactic responses to chemoeffector, chemotactic responses to chemoeffector, bacterial migration over time, rapid chemotactic response, chemotactic response, investigating chemoeffector kinetic, lasting chemoeffector gradient, chemoeffector kinetics in the setup, chemoeffector, microfluidic assay, chemoeffector gradient, chemotaxi, using chemoattractant, pathogenic bacterium xanthomonas campestris pv, genetic screen, suitable for genetic screen, forward genetic screen, assay, chemical gradient, various motility mechanism, environmental bacterium shewanella oneidensi, impaired strain

Disclaimer

Spotlight Video

The video below is a supplement with extra context and tips, as part of the protocols.io Spotlight series, featuring conversations with protocol authors.

https://www.youtube.com/embed/bKsg9UKT9kU?si=5Uow-v7Kf0WKEmA8

This protocol was peer reviewed and accepted for publication in Plos One protocol

Abstract

Bacteria use various motility mechanisms to explore their environments. Chemotaxis is the ability of a motile bacterial cell to direct its movement in response to chemical gradients. A number of methods have been developed and widely used to study chemotactic responses to chemoeffectors including capillary, agar plug, microscopic slide, and microfluidic assays. While valuable, these assays are primarily designed to monitor rapid chemotactic responses to chemoeffectors on a small scale, which poses challenges in collecting large quantities of attracted bacteria. Consequently, these setups are not ideal for experiments like forward genetic screens. To overcome this limitation, we developed the Large Scale Bacterial Attraction assay (LSBA), which relies on the use of a Nalgene™ Reusable Filter Unit and other materials commonly found in laboratories. We validate the LSBA by investigating chemoeffector kinetics in the setup and by using chemoattractants to quantify the chemotactic response of wild-type, and motility impaired strains of the plant pathogenic bacterium *Xanthomonas campestris* pv. *campestris* and the environmental bacterium *Shewanella oneidensis*. We show that the LSBA establishes a long lasting chemoeffector gradient, that the setup can be used to quantify bacterial migration over time and that the LSBA offers the possibility to collect high numbers of attracted bacteria, making it suitable for genetic screens.

Troubleshooting

Disclaimer

Spotlight Video

The video below is a supplement with extra context and tips, as part of the protocols.io Spotlight series, featuring conversations with protocol authors.

https://www.youtube.com/embed/bKsg9UKT9kU?si=5Uow-v7Kf0WKEmA8

This protocol was peer reviewed and accepted for publication in Plos One protocol

Files

🔍 SEARCH

Protocol

NAME

Protocol preparation of bacterial cells

VERSION 1

CREATED BY

Boulanger Alice
université Paul sabatier - toulouse III

OPEN →

Protocol

NAME

Protocol LSBA setup

VERSION 1

CREATED BY

Boulanger Alice
université Paul sabatier - toulouse III

OPEN →

Protocol references

1. Blanvillain S, Meyer D, Boulanger A, Lautier M, Guynet C, Denancé N, et al. Plant carbohydrate scavenging through tonB-dependent receptors: a feature shared by phytopathogenic and aquatic bacteria. PloS One. 2007;2: e224. doi:10.1371/journal.pone.0000224

2. Boyeldieu A, Poli J-P, Ali Chaouche A, Fierobe H-P, Giudici-Orticoni M-T, Méjean V, et al. Multiple detection of both attractants and repellents by the dCache-chemoreceptor SO_1056 of Shewanella oneidensis. FEBS J. 2022;289: 6752–6766. doi:10.1111/febs.16548

3. Cerutti A, Jauneau A, Auriac M-C, Lauber E, Martinez Y, Chiarenza S, et al. Immunity at Cauliflower Hydathodes Controls Systemic Infection by Xanthomonas campestris pv campestris. Plant Physiol. 2017;174: 700–716. doi:10.1104/pp.16.01852

4. Luneau JS, Baudin M, Quiroz Monnens T, Carrère S, Bouchez O, Jardinaud M-F, et al. Genome-wide identification of fitness determinants in the Xanthomonas campestris bacterial pathogen during early stages of plant infection. New Phytol. 2022. doi:10.1111/nph.18313

5. Dugé de Bernonville T, Noël LD, SanCristobal M, Danoun S, Becker A, Soreau P, et al. Transcriptional reprogramming and phenotypical changes associated with growth of Xanthomonas campestris pv. campestris in cabbage xylem sap. FEMS Microbiol Ecol. 2014;89: 527–541. doi:10.1111/1574-6941.12345