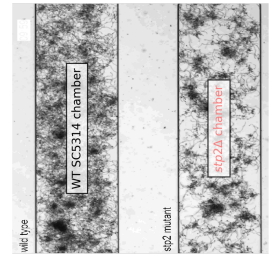Feb 06, 2020   Version 2

🌐 Bioflux Analyses V.2

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

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Keywords: Candida Albicans, Biofilm, Biofilm Formation, Growth Rate, Image Preprocessing, Edge Detection, ODE Model, Parameter Estimation

Abstract

Biofilm formation under shear flow conditions was monitored using the Bioflux1000 device (Fluxion Biosciences, Inc.). In short, Candida albicans overnight cultures were washed in pre-warmed RPMI medium. Cells were seeded for 2-5 sec from the outlet well into the channels of Bioflux1000 flow chambers, which were primed before with warm medium. The cells were allowed to adhere to the channels for 90 min without any flow, followed by removal of non-adherent cells by flowing fresh, pre-warmed RPMI medium for 5 sec. Shear flow was set for time series experiments over 24 h biofilm formation and images were captured every 20 min. Two channels were investigated in parallel having a 10 × magnification to allow a direct comparison between a mutant and a reference (wild type) strain. Image capturing and stacks to movies was performed using the MetaMorph® Software (Molecular Devices).

Source material provided as AVI files was converted into single TIFF images as well as data frames containing meta data annotations. The individual image contains two growth chambers (wild type and mutant) separated by four edge lines. Images were rotated automatically to vertical alignment in order to carry out an automated chamber detection and analysis. The mean pixel intensity (i. e. grey scale value; reflecting cell density) of the individual chamber was calculated and added into the respective data frame.

An ODE model reflecting the logistic growth as well as the lag phase was fitted to the individual experiments. Fitting was carried out by minimising a cost function (unweighted least-squares-based) using the *Nelder-Mead* algorithm. Growth rate time series generated from the fitted model were used to compare wild type and mutant regarding the maximum observed growth rates at their respective time points.

All computations were performed using the programming language python (version 3.6.9) and the additional packages numpy (version 1.16.2), opencv-python (version 4.1.1.26), pandas (version 0.25.0), scipy (version 1.3.1) and scikit-image (version 0.15.0).

Attachments

ZIP

Bioflux_Analyses_sam...

10.9MB

Attachments

Bioflux_Analyses_sa

Bioflux_Analyses_sam...

10.9MB

Files

🔍 SEARCH

Protocol

NAME

Bioflux Analyses: Image Preprocessing

VERSION 2

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Protocol

NAME

Bioflux Analyses: Modelling

VERSION 2

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