Dispersing Labyrinthulomycete Cell Aggregates by Sonication

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Abstract

Sonication procedure implemented to reduce labyrinthulomycete cell aggregation from culture for subsequent growth analyses.

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Protocol

Set up QSONICA Q800R Sonicator

Step 1.

Turn on QSONICA Q800R Sonicator. Add required distilled water and allow the water to reach a temperature of 4 C.

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Output frequency: 20KHz

Prepare cells

Step 2.

Pipette 1 - 1.2ml of cell culture into a 1.5ml eppindorf tube. Gently vortex. Invert the tube several times before placing in QSONICA adapter to maintain the cells in suspension.

Sonicate cells

Step 3.

Program QSONICA to sonicate for 1 minute at 100% amplitude followed by a 30 second intermission and another 1 minute at 100% amplitude. Put on necessary protective ear wear. Place the 1.5ml tubes of suspended cells into QSonica adapter and then into QSONICA. Close the lid of the machine and immediately begin sonication procedure by hitting START.

NOTES
Mariana Rius 30 Mar 2017

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Aurantiochytrium limacinum (ATCC MYA-1381)

Mass cell lysis was seen after 2 minutes of sonication at 100% amplitude

Schizochytrium aggregatum (ATCC 28209)

Can withstand up to 3 minutes sonication at 100% amplitude without significant cell lysis

Aplanochytrium stocchinoi

Can withstand 4 minutes at 100% amplitude with little to no visible cell lysis

Microscopy of post-sonicated cells

Step 4.

Identify cell mortality and remaining cell aggregation following sonication using microscopy and adjust sonication procedure as necessary.

This protocol has been successfully used to break up aggregates of *Aplanochyrium*, *Aurantiochytrium*, and *Schizochytrium* from cultures grown in liquid 790 medium.

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