

Shake Flask Cultivation

Introduction

Here, we describe the protocol for the set-up and execution of the shake-flask cultivations.

Media recipes, spore suspensions, sampling procedures and fluorescence measurements are covered elsewhere.

Materials

› Materials

- › Pyrex shake flasks (with baffles)
- › Minimal media
- › Spore suspension

› Consumables

- › Cotton stopper
- › Tin foil

Procedure

Preparations and inoculation

1. A breathable cotton ball was added to the mouth of a glass shake-flask (with baffles), covered with tin foil, and then autoclaved
2. 150 mL minimal media (see MEDIA) was added to each shake flask
3. The shake-flasks were inoculated with their respective spore solutions to a final concentration of 10^6 spores/mL
4. Shake flasks were incubated for 5 days as described below

Conditions

5. Shake flasks were incubated at 30 C at 150 rpm for 5 days
6. Samples were taken as regularly as described in Sampling procedures