Vial-scale Propane strain cultivation for GC

Introduction

Based on article created by Menon et al. (2015) and Kallio et al. (2014).

Materials

- T5 media
  - 12 g tryptone, 24 g yeast extract, 4 mL glycerol, 0,17 M KH2PO4, 0,72 M K2HPO4, 20 g glucose per litre
  - 0,5 mM IPTG when inducing

Procedure

Preparation

1. Overnight culture with 20 mL of LB, incubation at 38°C and 180 rpm
2. Inoculate with 10% (*v/v*) of the previous culture
3. 20ml inoculum in 200 mL of TB media, keeping it at 37°C and 230 rpm until the optical density reaches 0,5 (OD600).
4. Induce with IPTG to a final concentration of 0,5 mM.
5. Grow for 4 h at 30°C and 200 rpm to prepare samples for propane detection.

Cultivation in vials

6. Centrifuge 200 mL cell culture at 4000 rpm (21°C, 10 min) and discard the supernatant.
7. Resuspend the cells in 50 ml TB media with 0,5 mM IPTG.
8. Transfer 8 ml resuspended culture into 22 mL crimp sealed GC vial and incubate at room temperature and 180 rpm for 3 hours. Overall four vials.
9. Remove 1,0 mL headspace gas manually or automatically and inject into GC with a gas tight syringe.
10. **Four paralled individual reactions for each data point from the same main culture.**
11. **Additional oxygen can be supplemented into the culture headspace using a gas-tight syringe and injection needle through the vial septum.**