

Microscopy (Oracoli)

Materials

LB 0.1% Carbenicillin

- DH5 α -strain with pSB1A2-I13522 (pTet GFP)
- DH5 α -strain with pSB1A2-I13521 (pTet RFP)
- 1 ml syringe
- 10 ml syringe
- Syringe pump
- PTFE tubing (OD 1.6 mm, ID 0.8 mm)
- Microfluidic chip

Methods

1. Inoculate strains from glycerol stocks in 5 mL overnight culture
2. Dilute 1:1000 and grow until OD 0.4
3. Centrifuge @ 500 rpm
4. Resuspend in 15% of the initial volume
5. Draw in 1ml syringe
6. Plug into outlet tubing and load channels until every field is filled
7. Fill the 10 ml syringe with medium and plug it into the inlet tubing
8. Pump the medium at 20 μ l/h through the fluidics
9. Acquire images at every field in 40x magnification -> BF, GFP, RFP channels every 5 min for 20 h