The experiment design of 2018 OUC-China collaboration

1. PURPOSE

Measuring the fluorescence and OD600 of the target strain

2. Materials

a. LB medium (peptone 10g/L, yeast extract 5g/L, Nacl 10g/L)

b. M9 medium (12.8 g/L Na2HPO·4.7H2O, 3g/L KH2PO4, 0.5 g/L NaCl, 1.67g/L NH4Cl, 1 mM thiamine hydrochloride, 0.4% glucose, 2 mM MgSO4, 0.1 mM CaCl2)

c. IPTG (0.125mM)

d. 96-well plates (the picture below)

e. target strain and DH5α

3. Protocol

a. Take the glycerine bacteria plate (30-40μl is enough) and culture overnight.

b. Single colony is selected and cultured overnight in 50ml LB medium (150rpm, 37℃).

c. Take 1ml LB bacteria solution into 99ml M9 medium.

d. Add IPTG after 4h culture (200rpm，37℃).

e. Starting with the addition of IPTG, OD600 and fluorescence were measured every hour, and 8 sets of data were measured.

4. Attentions:

a. The bacteria added with IPTG, without IPTG, DH5, M9 medium should be measured.

b. At the time of measurement, 200μl was taken from the bottle each time in the perforated plate, and 3 repeated groups were taken each time for each bottle bacteria.

c. When selecting single colonies, if the bacteria solution is too thick and can be diluted appropriately, three single colonies should be selected for biological repetition.

d. Fluorescence (485nm excitation light and 520nm emission light) and OD600.