Screening

Time : 30 min

I. Principle

This technique is used for identification of the best enzyme candidates by fluorescence.

II. Material

- Colonies to be screened
- 96 well plates
- Chloramphénicol 35 µg/mL
- LB medium

III. Method

This experiment has to be done in sterile conditions (Bunsen burner or MSC) in order to avoid contaminations.

- In a 96 well plate :
  - Add 1 mL of LB and 1 µL chloramphenicol in each well
  - Subculture the colonies of interest on the plate, following a plate map
  - Incubate overnight at 37°C and 200-250 rpm

- In a new 96 well plate :
  - Add 900 µL of LB and 0,9 µL of chloramphenicol in each well
  - Add 100 µL of the cultures from the first plate following the same plate map
  - Incubate 1h at 37°C and 200 rpm

- In a new 96 well plate :
  - Add 30 µL of fructose 250 g/L in each well
  - Add 120 µL of each culture from the previous plate following the same plate map
  - Set the plate reader for a 10h screening at 587 nm for excitation and 610 nm for émission