Motility

Given the different viscosity of the medium and the placement of the colony on/in the medium, it is possible to test for presence or absent of twitching, swarming and swimming motilities.

Preparation of motility plates

Three kinds of plates are prepared each containing a different agar concentration (recipe for 1 plate):

- **Twitching 1.5 %:**
  - 19 mL melted ABT medium with 2 % agar
  - 6 mL ABT medium preheated to 50 °C
  - 0.625 mL 20 % glucose
  - 0.625 mL 20 % Cas-amino acids

- **Swarming: 0.53 %:**
  - 7 mL melted ABT medium with 2 % agar
  - 18 mL ABT medium preheated to 50 °C
  - 0.625 mL 20 % glucose
  - 0.625 mL 20 % Cas-amino acids

- **Swimming: 0.3 % for swimming:**
  - 4 mL melted ABT medium with 2 % agar
  - 21 mL ABT medium preheated to 50 °C
  - 0.625 mL 20 % glucose
  - 0.625 mL 20 % Cas-amino acids

Dry the plates for 5 min. at 37 °C before use

Protocol

1. Transfer a single colony from ON plates with a toothpick to a motility plate:
   a. Twitching: The colony is stabbed through the agar to the bottom of the plate.
   b. Swimming: The colony is deposited in the middle of the agar plate
   c. Swarming: The colony is placed on the surface of the agar plate
2. Twitching plates are incubated at 37 °C for 48 hours.
3. Swimming and swarming plates are incubated at 30 °C for 24 hours – do not turn the plates upside down, agar will fall off due to low agar concentration.

*TIP! In twitching PAO1 pilA can be used as a negative control and in swimming and swarming PAO1 fliF can be used as a negative control.*