

12-Well Plate In Vitro Biofilms

1. Grow up an overnight 3 ml planktonic culture.
2. Coat each well of plate with 10 ul serum, using a spreader.
3. Incubate at 30°C for 30 minutes, without shaking or parafilm.
4. Count cells from overnight culture and adjust to 1×10^6 in RPMI (Karen's).
5. Add 200 ul of adjusted cells to each well, making sure to coat the bottom, and incubate at 30°C for 30 minutes without shaking.
6. Remove media from wells and wash with 300 ul 1XPBS.
7. Add 200 ul fresh RPMI to each well, wrap plate in parafilm then foil, and incubate at 37°C with shaking at 50 RPM.
8. For harvesting, remove media from wells and wash with 300 ul RPMI.
Add 200 ul of 1XPBS to each well and scrape with a scraper. Transfer to an appropriate tube and store as needed.