THP-1 cell culture and differentiation

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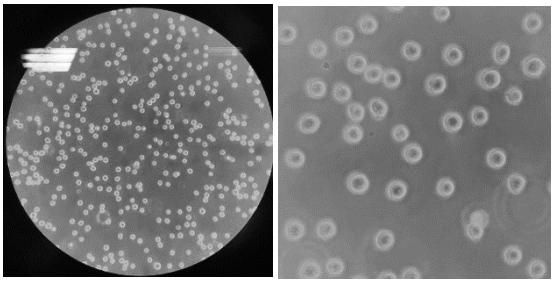
- **Materials**
  - Complete growth medium: RPMI-1640 Medium + 10 % FBS + 1% PS (Penicillin – Streptomycin) + 0.05 mM 2-mercaptoethanol (ATCC에서 권장, 추가하지 않아도 됨)
  - T-75 flasks

- **Process**

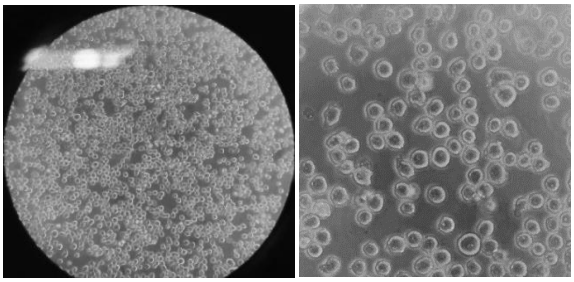
Step 1: Subculturing

- Resuspension: 4 x 10^6 cells 를 15mL complete growth media 에 띄워 T-75 flasks 에 넣음
- Subculture: 세포 농도가 ≒ 1.2 x 10^7 cells 까지 키움 (세포 농도가 1 x 10^6 cells/mL 를 초과하지 않게 함)
- Population doubling time (PDT): 약 26-28 hrs
- Medium Renewal: 3 days
- Culture Conditions: Atmosphere: air 95%; CO_2 5%
- Temperature: 37.0 ˚C

DAY 0



DAY 3



Step 2: Macrophage differentiation

- M0: PMA 100 ng/mL 24hr 분화 후, PMA 30 ng/mL 로 변경하여 48hr 분화 시킴
- M1: PMA 100 ng/mL 24hr 분화 후 PMA 30 ng/mL + LPS 20 ng/mL + hIFNɣ 20 ng/Ml 로 변경하여 48hr 분화 시킴
- M2: PMA 100 ng/mL 24hr 분화 후 PMA 30 ng/mL + hIL-4 20 ng/mL 로 변경하여 48hr 분화 시킴