**Ordering Information**

**HL-60** may be ordered from ATCC as a frozen ampoule.

- **Name:** HL-60, acute promyelocytic leukemia
- **ATCC #:** CCL-240

**Notes:**
This cell line grows in suspension.

**Materials List**

1. DMEM with 2mM L-glutamine (cellgro Cat# 10-013-CM)
2. Fetal Bovine Serum (cellgro Cat# 35-016-CV)
3. T75 & T225 culture flasks
4. Graduated pipets (1, 5, 25mL)
5. Penicillin-Streptomycin Solution (100X) (Cellgro Cat# 30-002-CI)
6. Hemocytometer
7. Micropipet w/ P20 tips
8. Microscope

**Growth Media for HL-60**

DMEM with 2mM L-glutamine (cellgro Cat# 10-013-CM)

20% FBS

1x Pen-Strep

**Procedure**

**A. Receipt of Frozen cells and starting cell cultures.**

1) Immediately place frozen cells in liquid nitrogen storage incubator.
2) Quickly thaw ampoule in 37°C water bath.
3) Transfer thawed cells to a T75 flask at 1 X 10^5 density in warm growth medium.
4) Allow cells to recover over night in 37°C, 5% CO2 humidified incubator.
5) Pour off medium the next day, replace with fresh medium and return to incubator.

**B. Sub-culture**

1) Propagate cells until density reaches ~8 X 10^5 (not to exceed 1 X 10^6).
2) Decant medium.
3) Wash cells with warm 1X PBS.
4) Immediately remove cells and pellet at 500 xg for 3 minutes (4°C)
5) Wash cells 2X with 1X PBS.
6) Gently re-suspend cell pellet in warm medium.
7) Seed at density of 1 X 10^5.
8) Record each subculture event as a passage.
C. **Maintenance**
   1) Change media the day after seeding and every 2-3 days thereafter.

D. **Harvest**
   1) Do not use cells that have been passed more than 25 times
   2) Remove cells from flasks according to protocol described above under 'sub-culturing'
   3) Examine viability using trypan blue staining.