

SOP: Propagation of K562
Date modified: 9/5/2008
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Ordering Information

K562 may be ordered from ATCC as a frozen ampoule.

Name: K562-chronic myelogenous leukemia
ATCC #: CCL-243

Notes:

This cell line grows in suspension.

Materials List

1. RPMI 1640 with 2mM L-glutamine (cellgro Cat# 10-040-CM)
2. Fetal Bovine Serum (cellgro Cat# 35-016-CV)
3. Sodium Pyruvate (cellgro Cat# 25-000-CI)
4. T225 & T25 culture flasks
5. Graduated pipets (1, 5, 25mL)
6. Penicillin-Streptomycin Solution (100X) (Cellgro Cat# 30-002-CI)
7. Hemocytometer
8. Micropipet w/ P20 tips
9. Microscope

Growth Media for K562

RPMI 1640 with 2mM L-glutamine
Sodium Pyruvate 10mM
10% FBS
Pen-Strep (1x)

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 T25 flask with 10ml of warm growth media.
- 4) Allow cells to recover over night in 37°C, 5% CO₂ humidified incubator.
- 5) The take cell count and spin down cells, 500g for 5 minutes, then decant old media
- 6) Re-suspend cells in warm fresh media at a volume to yield a density of 2x10⁵ cells/ml.

B. Sub-culture

- 1) Take cell counts every 48 to 72 hours.
- 2) Maintain cell density between 1x10⁵ and 1x10⁶ cells/ml.
- 3) Add fresh warm media when appropriate.
- 4) Record each subculture event as a passage.
- 5) Cells can be spun down, 500g for 5 minutes, rinsed with 1x PBS and re-suspend in a smaller volume of warm growth media when appropriate

C. Maintenance

- 1) Change media as cell density requires.

D. Harvest

- 1) Pass cells until desired cell number is achieved
- 2) Spin down cells and rinse with 1x PBS as described in `sub-culturing`