

K562 Cell Line

Catalog Number: PC-007 (For Research Use Only)

K562 cells were the first immortalized myelogenous leukemia line in humans. This cell line comes from a 53-year-old woman with chronic myelogenous leukemia. It has been shown that K562 can be multipotent hematopoietic malignant cells that spontaneously differentiate.

Product

One vial of 2 x 10^6 cells in Freezing Media. **IMPORTANT**: store the frozen cells in liquid nitrogen until you are ready to thaw and propagate them.

Handling cells upon arrival

It is strongly recommended that you propagate the cells by following instructions as soon as possible upon arrival.

Required Cell Culture Media

- Complete Growth Media In 450mL of RPMI, add 50mL FBS (10% final) and 5mL Penicillin/Streptomycin (1% final).
- **2x Freezing Media** Add 10% DMSO (final) to Complete Growth Media and sterile filter. Make fresh each time.

Materials required but not provided

(Can be substituted with a comparable third-party product)

- RPMI -- Hyclone P/N SH30027.01
- Fetal Bovine Serum (FBS) -- Fisherbrand P/N 03-600-511
- Penicillin/Streptomycin -- Hyclone P/N SV30010
- Trypsin *Hyclone P/N SH30236.02*
- Phosphate-buffered saline (PBS) -- Cellgro P/N 21-040-CV
- DMSO -- Sigma P/N D8418

Culture Procedure

- 1. Quickly thaw cells in a 37°C water bath with careful agitation. Remove from the bath as soon as the vial is thawed.
- Wipe the vial with 70% ethanol and transfer cell suspension into a 15ml centrifuge tube containing 9.0 mL warm complete culture medium.
- 3. Spin at 200 g, for 5minutes at room temperature.
- **4.** Resuspend cell pellet with the appropriate volume of complete medium and transfer the cell suspension into a T25 culture flask.
- 5. Place the flask with cells in a humidified incubator at 37° C with 5% CO₂.
- **6.** The next day, replace media with fresh Complete Growth Medium.
- 7. When cells are $6-8\times10^5$ cells/ml, split them 1:4 with fresh media. Add appropriate aliquots of the cell suspension to new culture vessels (T25 = 10 ml; T75 = 50 ml; T150 = 100 ml maximum volume). Grow cells to no more than 8 x 10⁵ cells/ml. Disperse clumps gently for counting.

Preparing frozen stocks

This procedure is designed for 100mm²dish or T75cm² flask. Scale volumes accordingly to other vessels.

- 1. When cells reach 90-100% confluency, freeze down cells.
- **2.** Transfer cells to a 15ml conical centrifuge tube and centrifuge at 200 g for 5 minutes to collect the cells into a pellet.
- **3.** Carefully aspirate the media and resuspend cells in 0.5mL complete growth media.
- **4.** Add 0.5mL of 2X freezing media and gently resuspend by pipetting up and down.
- 5. Transfer 1mL of cells into a cryogenic vial.
- 6. Place the cryogenic vial in a freezing container (*Nalgene # 5100-0001*) and store it at -80°C freezer overnight.
- 7. Transfer cells to liquid nitrogen for long-term storage.