Product description

The melan-p1 and -p2 cell lines are immortal melanocyte cell lines derived from neonatal Oca2p-cp/Oca2p-25H CH3 mice. These cell lines lack Oca2 (also referred to as p protein), a chloride ion channel found to relate melanosomal pH. Pathogenic variants in the human ortholog of Oca2, OCA2, result in oculocutaneous albinism type 2, a form of albinism characterized by variable hypopigmentation of the skin and hair and ocular defects. Individuals with OCA2 have significantly higher risk of developing skin cancers, including melanoma. These cells are hypopigmented despite the expression of tyrosinase. These cells are a useful model for OCA2 and for use in pigment cell biology studies. Melanoblast lines (melb-p3 and melb-p4) derived from pcp/p25H C3H mice are also available from CancerTools.org.

Name: Melan- p1 cell line

Organism: Mouse

Disease: Ocular Albinism

Tissue: Skin

Morphology: Small, oval-fusiform

Growth properties: Adherent

Model: Immortalised non-cancerous cell lines

Contributor(s)

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Properties

Product format: Frozen

Unpacking and storage:

1. Check all containers for leakage or breakage.

2. Remove the frozen cells from the dry ice packaging and immediately place the cells at a temperature below -130°C, preferably in liquid nitrogen vapor, until ready for use.

Recommended medium: RPMI 1640 + 10% FBS + 200 nM 12- O -tetradecanoy1 phorbol 13-acetate (TPA) + 200 pM Cholera toxin + 100 µM 2-Mercapto ethanol + 2 mM L-tyrosine

Seeding density: 2500-3000 cells/cm².

Culture conditions: 37.0°C ± 1.0°C incubator with 10.0% ± 1.0% CO_2

Cryopreservation medium: 7.5% v/v DMSO in complete cell culture medium. Freeze at a density of 1-3 x 10^6 cells/mL.

Handling instructions

1. Please ensure that vials are frozen when received, and store at **<-130 °C long term**. When removing frozen cells from storage, it is important to minimize exposure to room temperature (15 - 25°C). If not proceeding directly to thawing, place the cells on dry ice or in a liquid nitrogen container.
2. **Do not thaw at room temperature.** To thaw, swirl the vial quickly in a 37 °C water bath with O-ring and cap above the water to avoid contamination. Remove from the water bath with a small ice pellet remaining (this should not take more than 2 minutes) and wipe the exterior with 70% ethanol or isopropanol before transferring to a biosafety cabinet. Further steps should be conducted under aseptic conditions.
3. We strongly recommend that the volume of cell suspension is measured, and a 20 uL aliquot be set aside at this point for a viable cell count using trypan blue or similar dye.
8. Gently transfer the cell suspension to a 20 mL centrifuge tube. Dilute by gradually adding 20 mL of complete recommended medium. It is important to **dilute very slowly, dropwise** e.g. add first 1 mL with mixing over about 30 sec, second 1 mL over 10 sec, then remaining 18ml over 30 sec. (Note that freezing medium is highly hypertonic and DMSO needs time to diffuse out without damaging the cells).
9. Centrifuge the cell suspension at 100 x g for 5 minutes at room temperature (this time can be utilized to obtain the viable cell count from the aliquot).
10. Carefully remove the supernatant with a pipette, leaving a small amount of medium to ensure the cell pellet is not disturbed. Resuspend the cell pellet by gently flicking the tube.
11. Gently add required volume of culture medium and transfer to a suitable cell culture flask at the recommended density.
12. Subculture routine: Split sub-confluent cultures every 4-5 days. Incubate with 0.025% trypsin–0.02% EDTA at 37 °C for 5 minutes or until cells detach.

Note: It is important to work very quickly with this cell line post-thaw, as DMSO is especially toxic to pigmented cells.

References

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Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: Melan-p1 cell line, was invented by Elena Sviderskaya (CancerTools.org #162029).

Additional Resources

Protocols from the inventor lab: https://www.sgul.ac.uk/about/our-institutes/neuroscience-and-cell-biology-research-institute/genomics-cell-bank/materials-and-methods