

Product description

The Melan-a cell line is an immortal melanocyte cell line derived from embryonic mouse skin. It was the first established non-tumorigenic mouse melanocyte cell line and has become a widely used model for melanocytes studies. Melanocytes are pigment-producing cells located in the bottom layer of the skin's epidermis and in other tissues. Melan-a cells display the pigmentation and morphology typical of normal melanocytes, expressing both gp100 and the melanocyte marker Melan-A. They are diploid in chromosome number and syngeneic with C57BL mice and B16 melanoma sublines, facilitating comparative investigations. These cells are visibly pigmented, smaller than B16 melanoma cells and retain all tested characteristics of normal melanocytes, except senescence. Among retained features, they also display a proliferative response to cholera toxin in the presence of tetradecanoyl phorbol acetate (TPA). The availability of robust cellular models that reflect normal skin melanocytes, such as the Melan-a cell line, is essential for studying the etiology and abnormalities of skin cancers and melanomas, allowing comparisons at a molecular and cellular level.

Name: Melan-a cell line

Cancer: Skin cancer

Cancers detailed: Melanoma

Organism: Mouse

Tissue: Embryonic skin

Growth properties: Adherent

Model: Immortalised line; Non-tumorigenic in syngeneic and nude mice

CRISPR Edited: No

Biosafety level: 1

Cellosaurus id: CVCL_4624

Contributor(s)

Inventor: Dorothy Bennett

Institute: St George's University of London

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 vapour, until ready for use.

Recommended medium: RPMI-1640 + 200 nM phorbol-12-myristate-13-acetate (TPA)
+ 10% Foetal Bovine Serum (FBS)

Subculture: Passage at ~80-85% confluency. Carefully remove the medium from the T75 tissue culture flask containing the 80-85% confluent layer of Melan-a cells. Rinse the flask with 10 mL of 1X PBS. Aspirate after the rinse. Apply 5-7 mL of Accutase to the flask and incubate in a 37°C incubator for 3-5 minutes. Inspect the flask and ensure the complete detachment of cells by gently tapping the side of the flask with the palm of your hand. Add 5-7 mL of Melan-a growth medium to the flask. Gently rotate the flask to mix the cell suspension. Transfer the dissociated cells into a 15 mL conical tube. Centrifuge the tube at 300 x g for 3-5 minutes to pellet the cells. Discard the supernatant, then loosen the cell pellet by tapping the tip of the tube with a finger. Apply 2-5 mL of Melan-a growth medium into the tube and resuspend the cells thoroughly by pipetting up and down. **IMPORTANT: Do not vortex the cells.** Count the number of cells using a haemocytometer. Split and plate the cells into new flasks at the desired density (typical split ratio is 1:4 or 1:6).

Culture conditions: 37.0°C ± 1.0°C incubator with 5.0% ± 1.0% CO₂

Handling instructions

1. Prepare Melan-a growth medium: RPMI-1640, 10% FBS, and 200 nM phorbol-12-myristate-13-acetate (TPA), to thaw and expand the cells. The cells can grow on normal tissue culture-ware surfaces without any additional coating.
2. Remove the vial of frozen Melan-a cells from liquid nitrogen and thaw rapidly by incubating in a 37°C water bath. Closely monitor until the cells are completely thawed. Maximum cell viability is dependent on the rapid and complete thawing of frozen cells. **IMPORTANT: Do not vortex the cells.**
3. As soon as the cells are completely thawed, disinfect the outside of the vial with 70% ethanol. Proceed immediately to the next step.
4. In a microbiological safety cabinet, use a 1- or 2 mL pipette to transfer the cells into a sterile 15 mL conical tube. Be careful not to introduce any bubbles during the transfer process.
5. Using a 10 mL pipette, slowly add dropwise 9 mL of Melan-a growth medium into the 15 mL conical tube. **IMPORTANT: Do not add the entire volume of media all at once to the cells.** This may result in decreased cell viability due to osmotic shock.
6. Gently mix the cell suspension by slowly pipetting up and down twice. Be careful not to introduce any bubbles. **IMPORTANT: Do not vortex the cells.**
7. Centrifuge the tube at 300 x g for 2-3 minutes to pellet the cells.
8. Decant as much of the supernatant as possible. Steps 5-8 are necessary to remove any residual cryopreservative (DMSO).
9. Resuspend the cells in 15 mL of Melan-a growth medium.
10. Transfer the cell mixture into a T75 tissue culture flask.
11. Incubate the cells at 37°C in a humidified incubator with 5% CO₂.

References

- Gabel et al. 2024. Nat Commun. 15(959):1-14. PMID: 38302465.
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- Unapanta et al. 2023. J Biol Chem. 299(10):105192. PMID: 37625589.
- Schadendorf et. al. 2018. Nat Rev Dis Primers. 392(10151): 971-984. PMID: 30238891
- Bennett et al. 1987. Int J Cancer. 39(3):414-418. PMID: 3102392. Nilsen et al. 2016. Eur J Oral Sci. PMID: 27711994

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: Melan-a cell line, was invented by Dorothy Bennett (CancerTools.org #153599).

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