YUMM1.7D4 Mouse Melanoma Cell Line

Cancer Cell Line

Cat. # SCC227

Pack size: ≥1x10^6 viable cells/vial

Store in liquid nitrogen

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NOT FOR USE IN DIAGNOSTIC PROCEDURES.
NOT FOR HUMAN OR ANIMAL CONSUMPTION.
THIS PRODUCT CONTAINS GENETICALLY MODIFIED ORGANISMS

Data Sheet

Background

The great promise of immune-based therapies in cancer and recent progress in successful application of these approaches has brought to the fore the necessity of immune-competent models to evaluate immune system responses to cancer cells. Immunocompetent genetically-engineered mouse models harboring discrete genetic drivers of melanoma are essential for studying potential immunotherapies but are limited by the need to maintain colonies of multiple genotypes necessary to generate mouse models with appropriate genetic backgrounds.

The YUMM1.7D4 mouse melanoma cell line is a diploid clone of the YUMM1.7 cell line, developed as an immunocompetent cell line that recapitulates genetic drivers found in a significant proportion of human melanomas.[1] The diploid genotype of YUMM1.7D4 cells greatly enhances efficiency of CRISPR-based screens with higher frequency of biallelic knockout compared to tetraploid cells. YUMM1.7D4 is syngeneic with the immunocompetent C57/B1/6 mouse background, retaining genetic markers of the *Braf/Pten* mouse model in which melanoma tumors develop with short latency.[2] The genetic tractability of the YUMM1.7D4 mouse melanoma cell line is especially suited for evaluation of potential immunotherapies and represents a valuable tool for tumor immunology and cancer biology.

Source

The YUMM1.7D4 mouse melanoma cell line is a diploid clone derived from the YUMM1.7 cell line, which originated from a 4-hydroxytamoxifen-induced melanoma tumor in a male C57/B1/6 mouse into which mutations from the *Braf/Pten* genetically-engineered mouse model had been introduced via backcrossing. The YUMM1.7D4 cell line harbors the *Braf* V600E mutation and is homozygous negative for wild-type *Pten* and *Cdkn2*.[1]

Storage and Handling

YUMM1.7D4 mouse melanoma cell line should be stored in liquid nitrogen. The cells can be cultured for at least 10 passages after initial thawing without significantly affecting the cell marker expression and functionality.

Quality Control Testing

- Each vial contains ≥ $1X10^6$ viable cells.
- Cells are tested negative for infectious diseases by a Mouse Essential CLEAR panel by Charles River Animal Diagnostic Services.
- Cells are verified to be of mouse origin and negative for inter-species contamination from rat, chinese hamster, Golden Syrian hamster, human and non-human primate (NHP) as assessed by a Contamination CLEAR panel by Charles River Animal Diagnostic Services.
- Cells are negative for mycoplasma contamination.

Representative Data

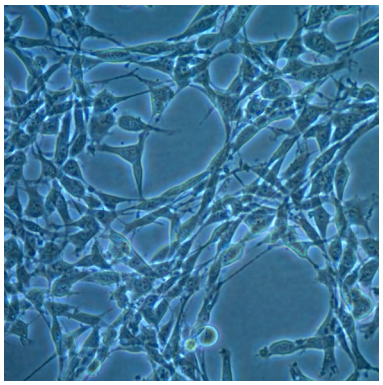

Figure 1. YUMM1.7D4 cells two days after thawing in a T75 flask (**A**).

References

1. Meeth K et al. (2016) The YUMM lines: a series of congenic mouse melanoma cell lines with defined genetic alterations. *Pigment Cell Melanoma Res* 29(5): 590-597.
2. Dankort D et al. (2009) Braf(V600E) cooperates with Pten loss to induce metastatic melanoma. *Nat Genet*. 41(5): 544-552.

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Protocols

Thawing Cells

1. Do not thaw the cells until the recommended medium is on hand. Cells can grow on normal tissue cultureware surfaces without any additional coating.

   YUMM1.7D4 Expansion Medium: Cells are thawed and expanded in DMEM/F12 medium (Cat. No.DF-042-B) supplemented with 10% FBS (Cat. No. ES-009-B) and 1X non-essential amino acids (Cat. No. TMS-001-C).
2. Remove the vial of frozen YUMM1.7D4 cells from liquid nitrogen and incubate 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 laminar flow hood, use a 1 or 2 mL pipette to transfer the cells to 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 YUMM1.7D4 Expansion Medium (Step 1 above) to 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 residual cryopreservative (DMSO).
9. Resuspend the cells in 15 mL of YUMM1.7D4 Expansion Medium.
10. Transfer the cell mixture to a T75 tissue culture flask.
11. Incubate the cells at 37°C in a humidified incubator with 5% CO_2.

Subculturing Cells

Note: Cells form a confluent monolayer that easily detach and may be difficult to resuspend in liquid culture.

1. Carefully remove the medium from the T75 tissue culture flask containing the confluent layer of YUMM1.7D4 cells.
2. Rinse the flask with 10 mL 1X PBS. Aspirate after the rinse.
3. Apply 5-7 mL of Accutase or trypsin-EDTA solution and incubate in a 37°C incubator for 3-5 minutes.
4. Inspect the flask and ensure the complete detachment of cells by gently tapping the side of the flask with the palm of your hand.
5. Add 5-7 mL of YUMM1.7D4 Expansion Medium to the plate.
6. Gently rotate the flask to mix the cell suspension. Transfer the dissociated cells to a 15 mL conical tube.
7. Centrifuge the tube at 300 x g for 3-5 minutes to pellet the cells.
8. Discard the supernatant, then loosen the cell pellet by tapping the tip of the tube with a finger.
9. Apply 2-5 mL of YUMM1.7D4 Expansion Medium to the conical tube and resuspend the cells thoroughly.

   IMPORTANT: Do not vortex the cells.
10. Count the number of cells using a hemocytometer.
11. Plate the cells to the desired density. Typical split ratio is 1:6.

Cryopreservation of Cells

YUMM1.7D4 Mouse Melanoma Cell Line may be frozen in the expansion medium plus 10% DMSO using a Nalgene slow freeze Mr. Frosty container.

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