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Data Sheet

YUMMER1G.1F Mouse Cell Line

Cancer Cell Line
Cat. # SCC246

pack size: ≥1x10^6 viable cells/vial

Store at: liquid nitrogen

FOR RESEARCH USE ONLY
NOT FOR USE IN DIAGNOSTIC PROCEDURES
NOT FOR HUMAN OR ANIMAL CONSUMPTION

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. Melanomas exhibit relatively high somatic mutation burden, and these mutations may act as neoantigens that generate anti-tumor immune responses. The development of immunocompetent cell models is critical to the advancement of cancer immunotherapy and understanding of immunity, although few tractable model systems are available.

The Yale University Mouse Melanoma (YUMM) lines are a comprehensive system of melanoma cell lines harboring defined genetic drivers of melanoma that are syngenic with the immunocompetent C57/B1/6 mouse model. YUMM cell lines were derived from the Braf/Pten genetically engineered mouse melanoma model, a highly penetrant GEMM in which melanomas develop with short latency. YUMM Exposed to Radiation (YUMMER) cell lines are clonal isolates of YUMM lines exposed to UVB radiation to reflect the high somatic mutation burden in melanomas. YUMMER cells harbor a high frequency of stable somatic mutations compared to YUMM cells; these mutations have been shown to stimulate host adaptive immune response.[3] The YUMMER1.G1F mouse melanoma cell line is both immunocompetent and reflective of the somatic mutations common in melanomas. YUMMER1.G1F carries three driver mutations of melanoma: *Braf* V600E, *Pten -/-* and *Cdkn2 -/-*.[1,2] YUMMER1.G1F cells are diploid, allowing for enhanced knockout frequency in CRISPR-based screens, are of female genotype, and have pheomelanin-producing background.[4] The characteristics of the YUMMER1.G1F cell line make it a valuable model for investigating sex-related differences in immune checkpoint inhibition and mechanisms of anti-tumor responses.

Source

The YUMMER1.G1F mouse melanoma cell line is a diploid clonal isolate from YUMMER.G cells exposed to UVB radiation.[4] The original YUMM1.1 cell line was derived from a 4-hydroxytamoxifen-induced melanoma tumor in a C57/B1/6 mouse into which mutations from the *Braf/Pten* genetically-engineered mouse model had been introduced via backcrossing.[1]

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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, human, chinese hamster, Golden Syrian hamster, and Non-human Primate (NHP) as assessed by a Contamination Clear panel by Charles River Animal Diagnostic Services
- Cells are negative for mycoplasma contamination.

Storage and Handling

YUMMER1G.1F cells should be stored in liquid nitrogen. The cells can be cultured for at least 10 passages after initial thawing without significantly affecting functionality.

Representative Data

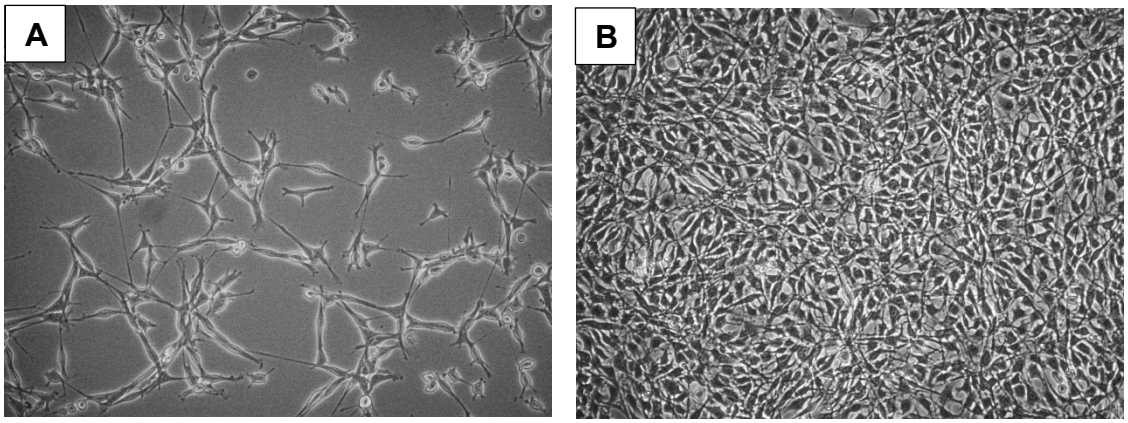

Figure 1. Bright-field images of YUMMER1G.1F one (A) and three (B) days after thaw in a T75 flask.

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.

   Cells are thawed and expanded in YUMMER1G.1F Expansion Medium comprised of DMEM/F12 (Sigma DF-041-B) with 1X Non-Essential Amino Acids (Sigma TMS-001-C), 10% FBS (Sigma ES-009-B) and 1X penicillin/streptomycin (Sigma TMS-AB2-C, optional).

2. Remove the vial of frozen YUMMER1G.1F 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.

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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 50 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 YUMMER1G.1F 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 YUMMER1G.1F 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

1. YUMMER1G.1F cells should be passaged at ~80-85% confluency. Do not allow the cells to grow over 85% confluency.
2. Carefully remove the medium from the T75 tissue culture flask containing the 80% confluent layer of YUMMER1G.1F cells.
3. Rinse the flask with 10 mL 1X PBS. Aspirate after the rinse.
4. Apply 5-7 mL of Accutase and incubate in a 37°C incubator for 3-5 minutes.
5. Inspect the flask and ensure the complete detachment of cells by gently tapping the side of the flask with the palm of your hand.
6. Add 10 mL of YUMMER1G.1F Expansion Medium to the plate.
7. Gently rotate the flask to mix the cell suspension. Transfer the dissociated cells to a 15 mL conical tube.
8. Centrifuge the tube at 300 x g for 3-5 minutes to pellet the cells.
9. Discard the supernatant, then loosen the cell pellet by tapping the tip of the tube with a finger.
10. Apply 2-5 mL of YUMMER1G.1F Expansion Medium to the conical tube and resuspend the cells thoroughly.

    IMPORTANT: Do not vortex the cells.
11. Count the number of cells using a hemocytometer.

Cryopreservation of Cells

YUMMER1G.1F cells may be frozen in YUMMER1G.1F Expansion Medium and 10% DMSO using a Nalgene slow freeze Mr. Frosty container.

References

1. *Pigment Cell Melanoma Res* 2016, 29(5): 590-597.
2. *Nat Genet* 2009, 41(5): 544-552.
3. *Pigment Cell Melanoma Res* 2017, 30(4): 428-435.
4. Ramseier JY, Charos A, Park K, Damsky W, Bosenberg MW (2019). *Cancer Res* 79(13 Suppl) Abstract #4622.

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12. Plate the cells to the desired density. Typical split ratio is 1:6.

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