

YUMMER1.7 H2B-GFP Mouse Melanoma Cell Line

Cancer Cell Line

Cat. # SCC245

Pack size: $\geq 1 \times 10^6$

viable cells/vial

Store in liquid nitrogen

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Data Sheet

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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 can 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 immune responses, although few tractable model systems are available.

The YUMMER1.7 H2B-GFP mouse melanoma cell line is both immunocompetent and reflective of the somatic mutations common in melanomas. The YUMMER1.7 H2B-GFP cell line carries three driver mutations of melanoma: *Braf* V600E, *Pten* -/- and *Cdkn2* -/-.^{1,2} In addition, the YUMMER1.7 H2B-GFP cell line harbors a high frequency of stable UV-induced somatic mutations which have been shown to stimulate host adaptive immune response.³ YUMMER1.7-H2B-GFP cells stably express GFP fused to histone H2B, allowing visualization of chromosomal dynamics and facilitating in vivo detection of YUMMER1.7 melanoma cells and YUMMER1.7-derived tumors. The unique features of the YUMMER1.7 H2B-GFP cell line make it a valuable model for studies of immune checkpoint inhibition and mechanisms of anti-tumor responses.

Source

The YUMMER1.7 H2B-GFP mouse melanoma cell line is a tetraploid clonal isolate derived from YUMM1.7 cells exposed to UVB radiation into which a histone H2B-GFP fusion has been stably introduced via transfection. The original YUMM1.7 cell line was derived 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.¹ The YUMMER1.7 H2B-GFP cell line harbors the *Braf* V600E mutation and is homozygous negative for wild-type *Pten* and *Cdkn2*.³

Storage and Handling

YUMMER1.7 H2B-GFP 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 $\geq 1 \times 10^6$ 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

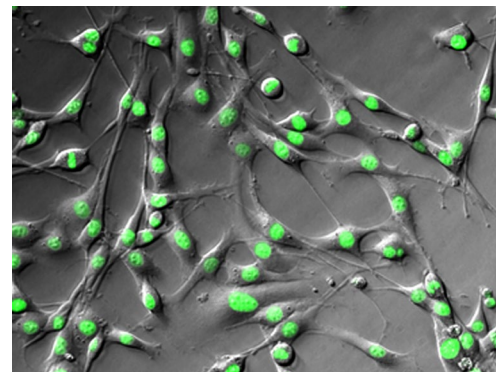


Figure 1. Overlay of bright-field and GFP fluorescent images.

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.
3. Wang J et al. (2017) UV-induced somatic mutations elicit a functional T cell response in the YUMMER1.7 mouse melanoma model. *Pigment Cell Melanoma Res* 30(4): 428-435.

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Protocol

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.

YUMMER1.7 H2B-GFP Expansion Medium: Cells are thawed and expanded in DMEM/F12 medium (Cat. No. DF-041-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 YUMMER1.7 H2B-GFP 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 YUMMER1.7 H2B-GFP 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 YUMMER1.7 H2B-GFP 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% CO₂.

Subculturing Cells

1. Carefully remove the medium from the T75 tissue culture flask containing the confluent layer of YUMMER1.7 H2B-GFP 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 YUMMER1.7 H2B-GFP 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 YUMMER1.7 H2B-GFP 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

YUMMER1.7 H2B-GFP 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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