AY-27 Rat Bladder Tumor Cell Line

Cancer Cell Line

Cat. # SCC254

Pack size: ≥1x10^6 viable cells/vial

Store in liquid nitrogen

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

Data Sheet

Background

Bladder cancer is one of the top ten most common malignancies worldwide, and no cure is available for metastatic stages of the disease.[1] Bladder cancer generally arises from the inner epithelial layer (urothelium).[1] A suitable bladder tumor model that recapitulates human disease both histologically and behaviorally is essential for evaluating new therapeutic agents and modes of treatment. Rat models more closely parallel human bladder carcinoma than mouse models regarding configuration and progress of the disease.[2]

The AY-27 rat bladder tumor cell line is a urothelium-derived transitional cell carcinoma[3] and is the most widely used animal model for bladder cancer. AY-27 cells are tumorigenic and are characterized by low adenylate cyclase activity,[4] lack of FasL expression, and positive staining for the urothelial marker cytokeratin-7.[5] Tumors derived from AY-27 cells demonstrate orthotopic growth, allowing exposure to anti-tumor drugs in a natural environment.[5] The high reproducibility and clinical relevance of the AY-27 tumor model has contributed greatly to the understanding of bladder cancer and evaluation of potential treatments.

Source

The AY-27 cell line was established from a primary bladder tumor induced by *N*-[4-(5-nitro-2-furyl)-2-thiazolyl]formamide (FANFT) treatment of Fischer F344 rats.[2]

Quality Control Testing

- Each vial contains ≥ $1X10^6$ viable cells.
- Cells are tested negative for infectious diseases by a Mouse/Rat Comprehensive CLEAR panel by Charles River Animal Diagnostic Services.
- Cells are verified to be of rat origin and negative for inter-species contamination from mouse, 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.

Storage and Handling

AY-17 Rat Bladder Tumor 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.

Representative Data

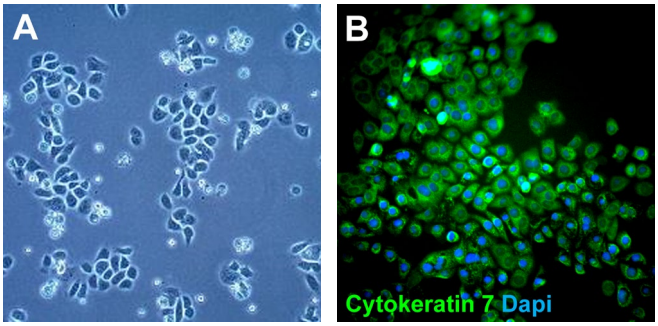

Figure 1. AY-27 cells one day after thawing in a T75 flask (A). Cells express cytokeratin-7 (B, CBL194F).

References

1. Sanli O et al. (2017) Bladder cancer. *Nat Rev Dis Primers* 3: 17022.
2. Oyasu R (1995) Epithelial tumors of the lower urinary tract in humans and rodents. *Food Chem Toxicol.* 33(9): 747-755.
3. Cohen SM et al. (1981) Transplantation and cell culture of rat urinary bladder carcinoma. *Invest Urol.* 19(3): 136-141.
4. Chlapowski FJ, Nemecek GM (1985) Aberrant cyclic adenosine 3':5'-monophosphate metabolism in cultures of tumorigenic rat urothelium. Cancer Res. 45(1): 122-127.
5. Xiao Z et al. (1999) Characterization of a novel transplantable orthotopic rat bladder transitional cell tumor model. Br J Cancer 81(4): 638-646.

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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.

   AY-27 Expansion Medium: Cells are thawed and expanded in RPMI-1640 (Sigma Cat. No. R0883) supplemented with 5-10% FBS (Cat. No. ES-009-B).
2. Remove the vial of frozen AY-27 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 AY-27 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 AY-27 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. Carefully remove the medium from the T75 tissue culture flask containing the confluent layer of AY-27 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 AY-27 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 AY-27 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

AY-27 Rat Bladder Tumor Cell Line may be frozen in the expansion medium plus 10% DMSO using a Nalgene slow freeze Mr. Frosty container.

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