ID8 Mouse Ovarian Surface Epithelial Cell Line

Immortalized Cell Line

Cat. # SCC145

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

Pack size: ≥1x10^6 viable cells/vial

Store in liquid nitrogen

Data Sheet

Background:

Ovarian cancer is the fourth leading cause of cancer-related deaths in women. The disease is frequently diagnosed at later stages, with tumors detected throughout the peritoneal cavity. Approximately 90% of ovarian tumors arise from ovarian surface epithelial cells[2]. Human and mouse ovarian surface epithelial cells (OSE) have been isolated and are used to develop ovarian cancer models. These models typically involve injection of the human/mouse OSE cells subcutaneously, intraperitoneally or orthotopically into immune deficient mice. A common drawback to these models is the absence of an intact immune system in the host mice.

In 2000, an immune-competent syngeneic mouse model for ovarian cancer was reported[1]. Mouse ovarian surface epithelial cells (MOSEC) isolated from C57BL/6 mice were found to spontaneously transform into malignant tumorigenic cells following prolonged passages (>20) in vitro[1]. Late passage (>20) MOSEC lost the classical "cobblestone" contact-inhibited in vitro properties reminiscent of normal epithelial cells and instead grew as multi-layered cell clusters indicative of transformed cells. Intraperitoneal injection of late passaged MOSEC into athymic and normal, immune-intact, syngeneic C57BL/6 mice gave rise to tumors throughout the abdominal cavity like those observed in women with Stage III and IV cancer[1]. MOSEC are thus useful syngeneic mouse models to study the role of the immune system in the establishment and progression of ovarian cancer.

ID8 is one of 10 clonal lines established from late passaged C57BL/6 murine ovarian surface epithelial cells (MOSEC)[1]. Intraperitoneal injection of each of the 10 clonal lines into C57BL/6 mice resulted in formation of peritoneal tumors and ascitic fluid. Of the 10 clonal lines, ID8 exhibited the highest tumor load. ID8 cell line is a highly published and well characterized cell line and is frequently used as a syngeneic mouse model for ovarian cancer.

Storage and Handling

ID8 Mouse Ovarian Epithelial Cells 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.

References

1. *Carcinogenesis* 2000; 21(4): 585-91.
2. *Gynecol Oncol* 2008; 108(2): 385-94.

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

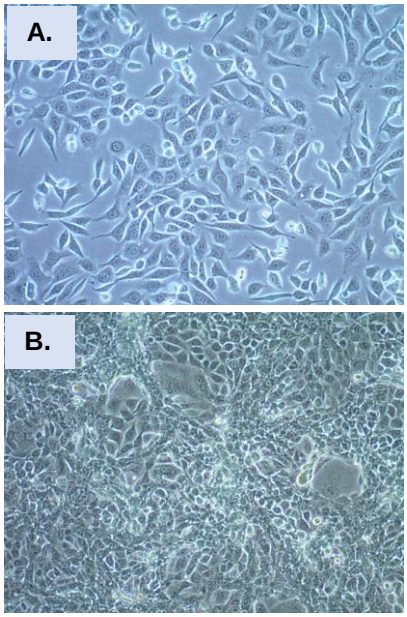

Figure 1. ID8 Mouse Ovarian Surface Epithelial Cell one (A) and two (B) days after thawing in a T75 flask. Late passage cells lose contact inhibition of growth and grew as multi-layered cells.

Rev.1.0/2018-02-23/SCC145/AN

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 High Glucose DMEM (Sigma Cat. No. D6429), 4% FBS (Cat. No. ES-009-B), 5 µg/mL insulin, 5 µg/mL transferrin and 5 ng/mL sodium selenite (1X ITS; Sigma Cat. No. I3146) and 1X Penicillin-Streptomycin Solution (Cat. No. TMS-AB2-C) (optional).
2. Remove the vial of frozen ID8 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 ID8 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 45 – 50 mL of ID8 Expansion Medium.
10. Transfer the cell mixture to a **T225** tissue culture flask.

    Note: *ID8 cells proliferate extremely rapidly. Do not thaw into a T75 flask. From time of thaw into a T225 flask, cells would be ready for passaging by day 2.*
11. Incubate the cells at 37°C in a humidified incubator with 5% CO_2.

Subculturing Cells

1. Carefully remove the medium from the T225 tissue culture flask containing the confluent layer of ID8 cells.
2. Rinse the T225 flask twice with 20 mL 1X PBS. Aspirate after each rinse.
3. Apply 10 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 12 mL of ID8 Expansion Medium to the plate.
6. Gently rotate the flask to mix the cell suspension. Transfer the dissociated cells to a 50 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 ID8 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 1:10-1:20). Cells proliferate extremely rapidly.

Cryopreservation of Cells

ID8 Mouse Ovarian Surface Epithelial Cell Line may be frozen in the expansion medium plus 10% DMSO using a Nalgene slow freeze Mr. Frosty container.

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