Data Sheet

UM-SCC-47 Squamous Carcinoma Cell Line

Cancer Cell line

SCC071

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.

Background

Head and neck squamous-cell carcinoma (HNSCC) is the 6th most common type of cancer world-wide. The cancer may occur in the lip, oral cavity, nasal cavity, paranasal sinuses, salivary glands, pharynx and larynx. Risk factors include smoking, alcohol consumption, betel nut chewing, wood dust exposures and human papilloma virus (HPV) infections. Approximately 15% of HNSCC contain genomic DNA from HPV.[2] In particular, HPV-16 occurs in 90-95% of all HPV-positive HNSCC cases.[2] In oral and pharynx cancer, HPV DNA is found in the tonsils in 45-67% of the cases. HPV DNA is found in 13-25% of the cases in the hypopharynx, 12- 18% in the oral cavity and 3-7% in the larynx.[2]

UM-SCC-47 is a unique head and neck squamous carcinoma cell line isolated from the primary tumor of the lateral tongue of a male[1] patient. The cell line contains 15-18 copies of integrated HPV-16 and approximately 10% of ALDH+ cancer stem cells.

Source

UM-SCC-47 was established at the University of Michigan[1] with written informed consent obtained from the patient and with the approval of the study by the Medical School Institutional Review Board as described by Brenner et al.

Short Tandem Repeat

D3S1358: 15	D16S539: 8, 13
TH01: 7, 9.3	CSF1P0: 11, 13
D21S11: 29, 30	Penta D: 9, 10
D18S51: 18	vWA: 18
Penta E: 12, 13	D8S1179: 15
D5S818: 11, 12	TPOX: 10, 11
D13S317: 8, 11	FGA: 23, 25
D7S820: 11	Amelogenin: X, Y

Cancer cell lines are inherently genetically unstable. Genetic instability may arise in the form of loss of heterozygosity of alleles at one or more genetic sites with increased passages.

Quality Control Testing

- Each vial contains ≥ $1x10^6$ viable cells.
- Cells are tested by PCR and are positive for HPV-16 and negative for Hepatitis A, B, C and HIV-1 and -2 viruses.
- Cells are negative for mycoplasma contamination.
- Each lot of cells are genotyped by STR analysis to verify the unique identity of the cell line.

Important Note

UM-SCC cell lines were derived in the lab of Dr. Thomas Carey at the University of Michigan and are exclusively distributed by us. PURCHASER may not distribute UM-SCC cells or derivatives to third parties.

Storage and Handling

UM-SCC-47 cells should be stored in liquid nitrogen. The cells can be cultured for at least 10 passages after the initial thaw without significantly affecting the cell marker expression and functionality.

Representative Data

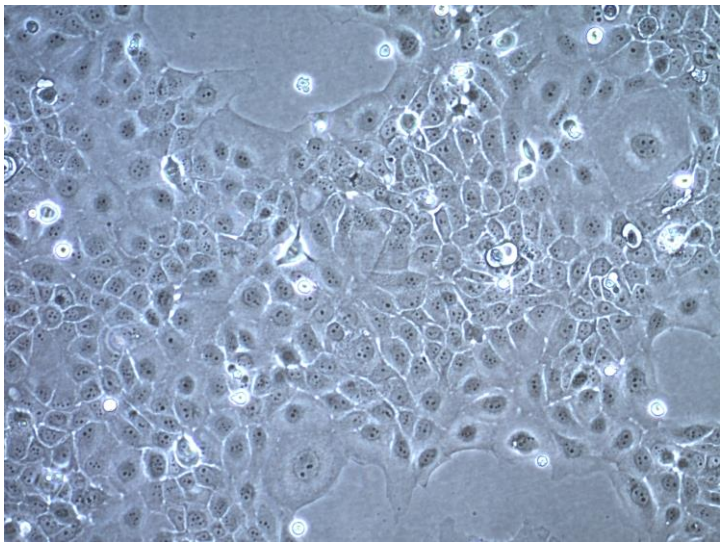

Figure 1: Day 2 after thaw.

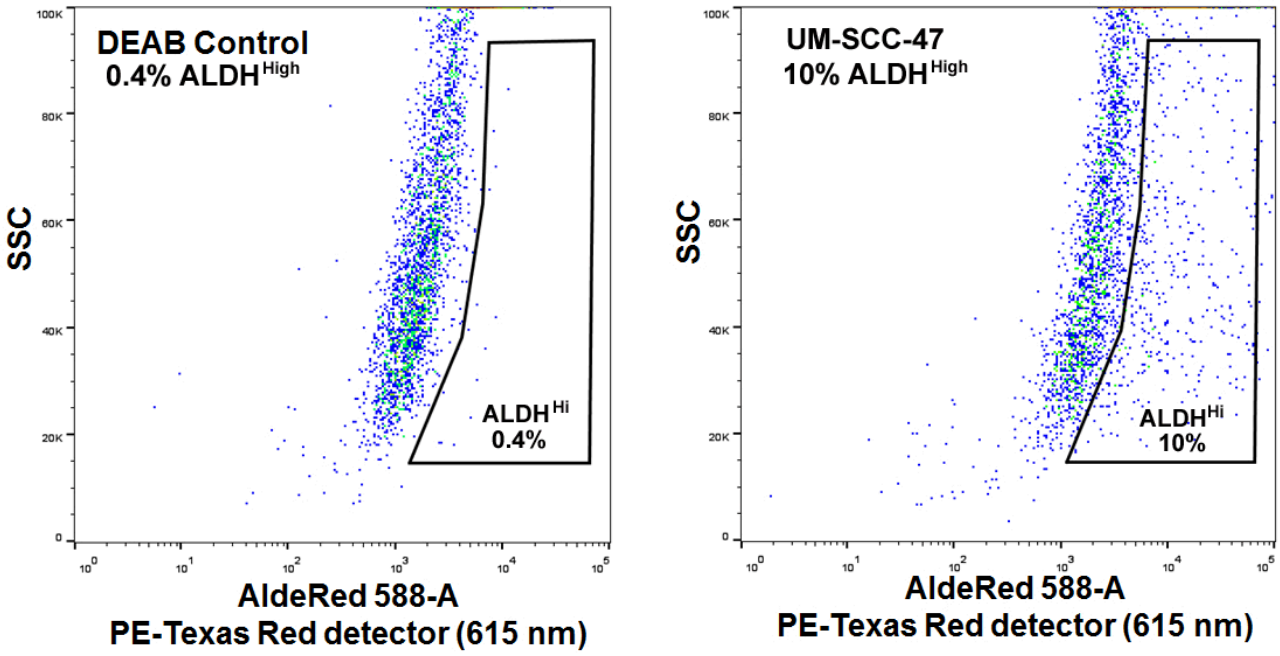

Figure 2: 10% ALDH+ cancer stem cells detected in UM-SCC-47 cells by AldeRed® 588-A (SCR150)

Protocols

Thawing of Cells

1. Do not thaw the cells until the recommended medium is on hand. Cells can grow on normal tissue culture ware surfaces without any additional coating. Cells are thawed and expanded in DMEM High Glucose (D5796), containing 10% FBS (ES009-B) and Non-Essential Amino Acids (TMS-001-C).
2. Remove the vial of frozen UM-SCC-47 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 10% FBS media (Step 1 above; pre-warmed to 37 °C) 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 a total volume of 10% FBS medium (pre-warmed to 37 °C).

10. Plate the cell mixture into a T75 tissue culture flask.
11. Incubate the cells at 37 °C in a humidified incubator with 5% CO_2.
12. The next day, exchange the medium with fresh 10% FBS media (Step 1 above) pre-warmed to 37 °C. Exchange with fresh medium every two to three days thereafter.
13. When the cells are approximately 90% confluent, they can be dissociated with Accutase® (SCR005) or trypsin (SM-2003-C) and further passaged or, alternatively, frozen for later use.

Subculturing of Cells

1. Carefully remove the medium from the T75 tissue culture flask containing the confluent layer of UM-SCC-47 cells.
2. Apply 3-5 mL of Accutase® or trypsin solution and incubate in a 37 °C incubator for 3-5 minutes.
3. Inspect the flask and ensure the complete detachment of cells by gently tapping the side of the flask with the palm of your hand.
4. Add 8 mL of 10% FBS medium (pre-warmed to 37 °C) to the plate.
5. Gently rotate the flask to mix the cell suspension. Transfer the dissociated cells to a 15 mL conical tube.
6. Centrifuge the tube at 300 x *g* for 3-5 minutes to pellet the cells.
7. Discard the supernatant, then loosen the cell pellet by tapping the tip pf the tube with a finger.
8. Apply 2 mL of 10% FBS media (pre-warmed to 37 °C) to the conical tube and resuspend the cells thoroughly.

   Important: Do not vortex the cells.
9. Count the number of cells using a hemocytometer.
10. Plate the cells to the desired density (typical split ratio is 1:3 to 1:6).

Cryopreservation of Cells

The UM-SCC-47 cells can be frozen in the expansion media plus 10% DMSO using a Nalgene® slow freeze Mr. Frosty® container.

References

1. Brenner, J.C., Graham, M.P., Kumar, B., Saunders, L.M.,Kupfer, R., Lyons, R.H., Bradford, C.R., Carey, T.E. (2010) Genotyping of 73 UM-SCC head and neck squamous cell carcinoma cell lines. Head Neck 34 (4):417-26.
2. Perez-Ordoñez, B., Beauchemin, M., and Jordan, R.C. (2006) Molecular biology of squamous cell carcinoma of the head and neck. J. Clin Pathol 59(5): 445-453.

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