

Data Sheet

SCCOHT-1 Human Small Cell Carcinoma Ovarian Cell Line

SCC437**Pack Size: $\geq 1 \times 10^6$ viable cells/vial****Store in liquid nitrogen.****FOR RESEARCH USE ONLY****Not for use in diagnostic procedures. Not for human or animal consumption.**

Background

Small cell carcinoma of the ovary hypercalcemic type (SCCOHT) is a rare form of aggressive cancer that primarily affects young women between the ages of 13 and 35.² Very few cell lines or other models are currently available to model this cancer. Although this cancer is often diagnosed at early stages, it has a very poor prognosis with more than half of patients diagnosed with stage IA dying within two years. This type of cancer has been distinguished from other related malignancies including transitional cell carcinoma of the ovary, ovarian epithelial tumors, and ovarian germ cell tumors.¹ Many other cell lines are available to study different ovarian carcinoma types, but no cellular model appears to exhibit the unique properties of SCCOHT-1.

Biopsy material from a recurrent SCCOHT tumor was used for explant culturing. The new culture became adherent and continued to proliferate. The newly established cell line was labeled "SCCOHT-1." Characterization analysis of the SCCOHT-1 cell line reveals an average diameter of $\sim 13\mu\text{m}$ and an estimated 36 hour doubling time. SCCOHT-1 cells are positive for CD29 and negative for CD24 and CD45 as assessed by flow cytometry. Injection of distinct subpopulations of the SCCOHT-1 cell line exhibited tumor development 100% of the time in NOD/SCID mice.¹ Additionally, research groups have identified SMARCA4 mutations resulting in loss of SMARCA4 protein which appears to be highly specific for SCCOHTs.² SMARCA4 protein forms an important ATPase of the SWI/SNF chromatin remodeling complex which is also often implicated as a tumor suppressor.² SCCOHT-1 cells provide a new tool to study the rare SCCOHT form of cancer and to develop relevant therapeutic strategies in the future.

Source

SCCOHT-1 cell line was created through explant culturing of biopsied recurrent SCCOHT tumor taken from a 31-year old female patient.¹

Short Tandem Repeat

D3S1358: 15, 17	D7S820: 10, 11	vWA: 14, 16	FGA: 25, 26	D8S1179: 13, 14
D21S11: 28, 31.2	D18S51: 13	D5S818: 11, 12	D13S317: 8, 12	D16S539: 9, 13
TH01: 7, 8	TPOX: 8, 9	CSF1P0: 10, 11	AMEL: X	Penta D: 12, 15
Penta E: 10, 13				

Quality Control Testing

- SCCOHT-1 cells are verified to be of human origin and negative for mouse, rat, Chinese hamster, Golden Syrian hamster, and non-human primate interspecies contamination, as assessed by a Contamination Clear panel by Charles River Animal Diagnostic Services.
- Cells tested negative for infectious diseases against a Human Essential CLEAR panel by Charles River Animal Diagnostic Services.
- Cells tested negative for mycoplasma.

Storage and Handling

SCCOHT-1 cells should be stored in liquid nitrogen until use. The cells can be cultured for at least 10 passages after initial thawing without significantly affecting the cell marker expression and functionality.

Representative Data

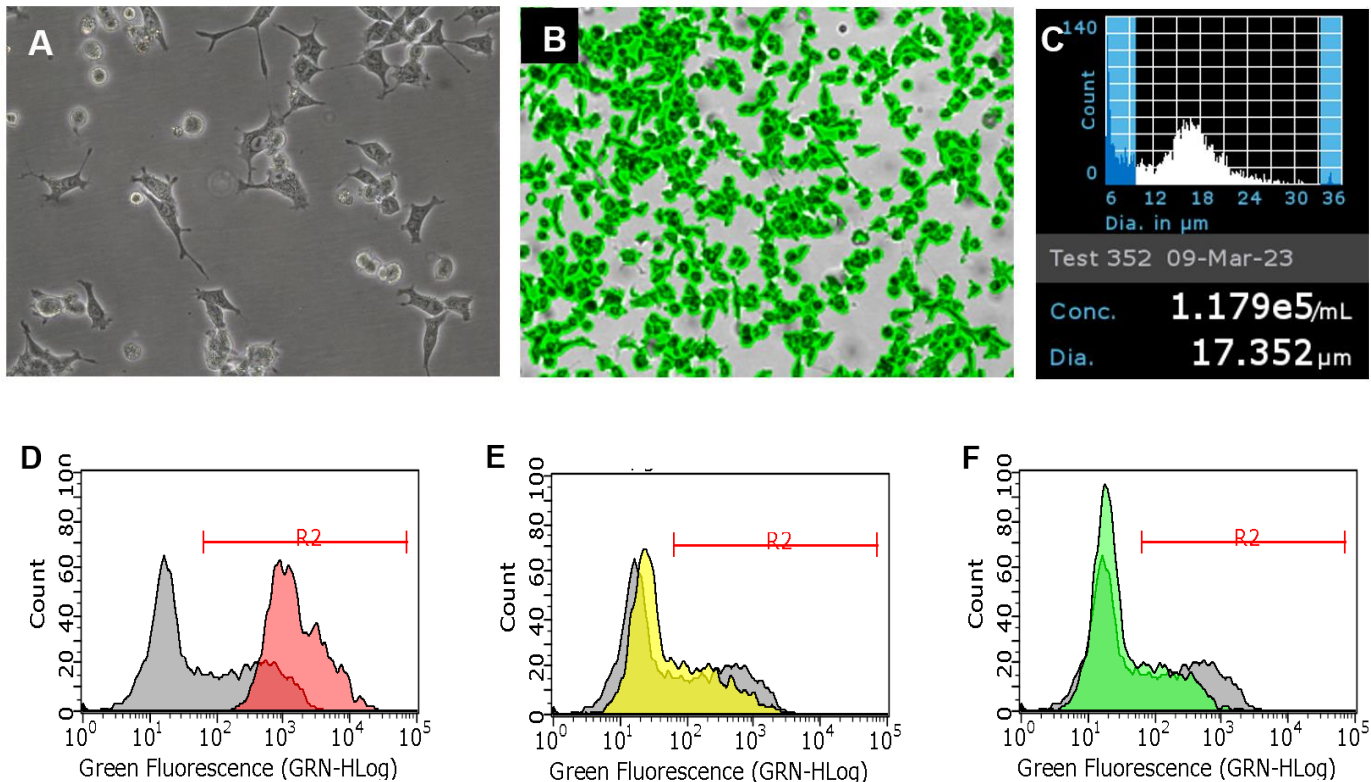


Figure 1. Brightfield image of SCCOHT-1 cells two days after thaw in a T75 flask (**A**). Cell confluency was assessed throughout the culture using the Millicell® Digital Cell Imager (**B**, Cat. No. MDCI10000). Cell counting was performed using Scepter™ 3.0 handheld automated cell counter using 60 μm sensors (**C**, Cat. No. PHCC360KIT). Cells express CD29 (**D**, Cat. No. SAB4700394). SCCOHT-1 cells show no significant expression of CD24 (**E**, Cat. No. CBL561), and CD45 (**F**, Cat. No. SAB4700471).

Protocols

Thawing the Cells

SCCOHT-1 cells are highly sensitive to cell thawing. There may be lower cell viability than expected during initial cell thaw, but cells will recover and proliferate rapidly.

1. Do not thaw the cells until the recommended medium is on hand. Cells can grow on standard tissue cultureware surfaces without any additional coating. Cells are thawed and expanded in SCCOHT-1 Expansion Medium comprising RPMI-1640 (Cat. No. R8758) containing 10% FBS (Cat. No. ES-009-B), and 2 mM L-Glutamine (Cat. No. TMS-002-C).
2. Remove the vial of frozen SCCOHT-1 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 SCCOHT-1 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 SCCOHT-1 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 the Cells

SCCOHT-1 cells are weakly adherent with many healthy cells growing as suspension cells. Both adherent and suspension cell populations should be collected and combined during passaging.

1. Do not allow the cells to grow to confluency. SCCOHT-1 cells should be passaged at ~80-85% confluency.
2. Carefully remove the medium from the T75 tissue culture flask containing the 80% confluent layer of SCCOHT-1 cells.
3. Rinse the flask with 10 mL 1X PBS. Aspirate after the rinse.
4. Apply 3-5 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 5-7 mL of SCCOHT-1 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 SCCOHT-1 Expansion Medium to the conical tube and resuspend the cells thoroughly. Large cell clumps may be broken up by gentle trituration.

IMPORTANT: Do not vortex the cells.

11. Count the number of cells using a hemocytometer or a Scepter™ 3.0 handheld automated cell counter.
12. Plate the cells to the desired density. Typical split ratio is 1:6.

Cryopreservation of the Cells

SCCOHT-1 cells may be frozen in SCCOHT-1 Expansion Medium supplemented with 10% DMSO using a Nalgene® slow freeze Mr. Frosty™ container.

References

1. Otte A, Göhring G, Steinemann D, Schlegelberger B, Groos S, Länger F, Kreipe H-H, Schambach A, Neumann T, Hillemanns P, et al. 2012. A tumor-derived population (SCCOHT-1) as cellular model for a small cell ovarian carcinoma of the hypercalcemic type. *Int J Oncol.* 41(2): 765-775.
2. Karnezis AN, Wang Y, Ramos P, Hendricks WP, Oliva E, D'Angelo E, Prat J, Nucci MR, Nielsen TO, Chow C. 2016. Dual loss of the SWI/SNF complex ATPases SMARCA4/BRG1 and SMARCA2/BRM is highly sensitive and specific for small cell carcinoma of the ovary, hypercalcaemic type. *J Pathol.* 238(3): 389-400.

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