Data Sheet

OCUG-1 Human Gallbladder Carcinoma Cell Line

SCC642

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

OCUG-1 Human Gallbladder Carcinoma Cell Line (SCC642), the human gallbladder carcinoma cell line, was isolated from the peritoneal effusion of a Japanese male patient with malignant gallbladder carcinoma. Although the primary tumor was diagnosed as a poorly differentiated adenocarcinoma, and the reconstituted tumors in nude mice also exhibited the same histologic characteristics, the OCUG-1 cells were found to abundantly express the Tumor-Associated Antigen 4 (TA-4), which had been identified as a serum marker for cervical squamous cell carcinoma.[2] This led Yamada *et al*. to propose that OCUG-1 might be a transitional form between adenocarcinoma and squamous cell carcinoma and represented a useful model system for investigating the squamatization processes.[1] Subsequently, one of the major components of TA-4, the Squamous Cell Carcinoma Antigen 1 (SCCA1, also known as SERPINB3, the Serine Protease Inhibitor Clade B Member 3), was shown to induce the squamatization and disease progression in pancreatic cancer by the inhibition of the degradation of the MYC proto-oncogene.[3]

As one of the relatively few available gallbladder cancer cell lines, OCUG-1 has been examined as a representative hapatobiliary cancer in genome-wide mutation and expression studies.[4,5] In one such study, Scherer *et al*. noted that the expression profile of the known primary gallbladder tumor markers is not commonly shared by the gallbladder cancer cell lines and suggested that investigators choose their cellular platforms based not only on the tissue origin of the tumors but also on the driver gene expression patterns underlying the specific mechanisms of tumorigenesis.[4]

Source

The OCUG-1 Human Gallbladder Carcinoma Cell Line (SCC642) cell line was isolated from the peritoneal effusion of a 43-year-old Japanese male patient with malignant gallbladder carcinoma.[1]

Short Tandem Repeat

D3S1358: 15	D7S820: 10, 11	vWA: 16	FGA: 21, 24	D8S1179: 14, 15
D21S11: 28, 29	D18S51: 16, 19	D5S818: 10, 12	D13S317: 8, 11	D16S539: 9, 13
TH01: 7, 9.3	TPOX: 8, 11	CSF1PO: 10, 12	AMEL: X	Penta D: 9
Penta E: 5, 20	Mouse: NA			

Quality Control Testing

- The OCUG-1 Human Gallbladder Carcinoma Cell Line (SCC642) 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

The OCUG-1 Human Gallbladder Carcinoma Cell Line (SCC642) 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

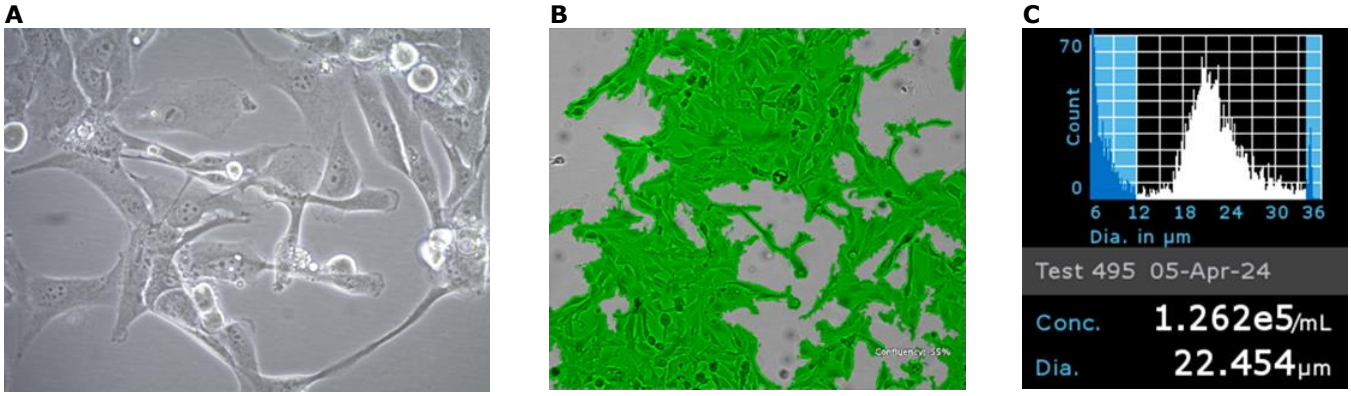

Figure 1. (**A**) Bright-field image of OCUG-1 cells a day after thaw in a T175 flask. (**B**) Cell confluency was assessed throughout the culture using the Millicell® Digital Cell Imager (MDCI10000). (**C**) Cell counting was performed using Scepter™ 3.0 Handheld Automated Cell Counter using 60 µm sensor tips (PHCC360KIT).

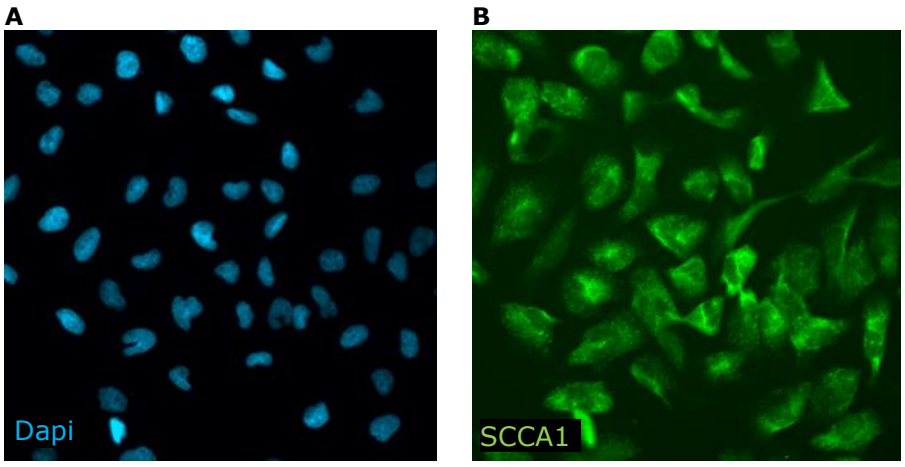

Figure 2. OCUG-1 cells express the Squamous Cell Carcinoma Antigen 1 (SCCA1). (**A**) OCUG-1 cells were labeled with DAPI (MBD0015) and (**B**) anti-SCCA1 antibodies (HPA055992).

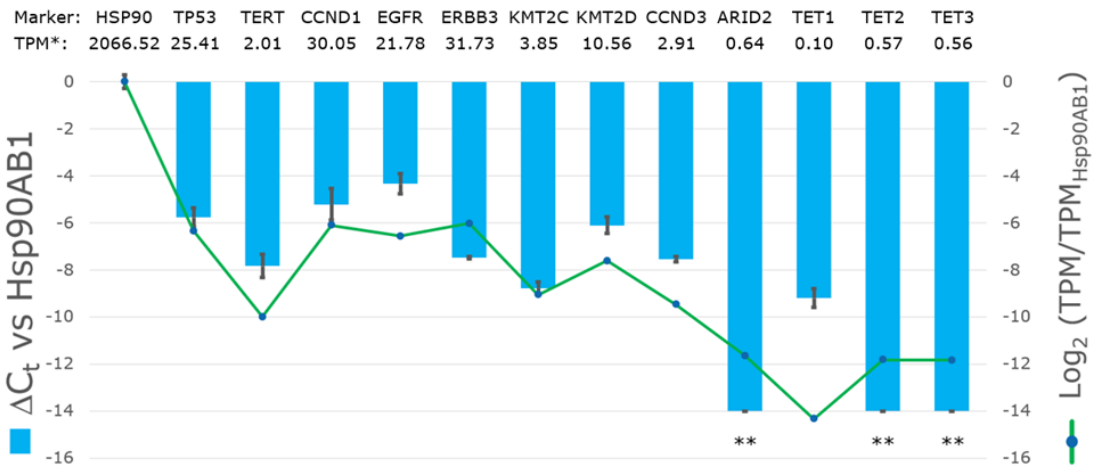

Figure 3. The Gallbladder carcinoma marker expression profile of OCUG-1. The expression levels of the 12 genes reported to be altered in at least 10% of primary GBC tumors from Japanese patients [4,6] were analyzed by RT-qPCR and expressed relative to that of Hsp90AB01 as the reference (ΔC_t, blue columns). The cDNA was prepared from 100 ng of total RNA with the Taqman RT Enzyme Mix (ThermoFisher Scientific, 4392728) using 500 nM gene-specific primers and amplified with the Fast SYBR Green Master Mix (ThermoFisher Scientific, 4385612) in the same tube. Each column represents the average of the triplicate measurements and the standard deviation. The qPCR primers used in this data sheet are listed in Table 1. The qPCR results were compared to the Log_2 of the transcripts-per-million numbers for the same genes (*) divided by that of Hsp90AB01 in the RNA-seq study reported by Scherer *et al*.[4] (green line). The expression of ARID2, TET2, and TET3 was not detectable by qPCR (**).

Table 1. RT-qPCR primers used in this data sheet

Target	Accession	Supplier	Part number	Sequence
Hsp90AB1	NM_007355.4	MilliporeSigma	Custom DNA synthesis	5′-AGTGTTGGGACTGTCTGG-3′ 5′-CACTTCCTCAGGCATCTTG-3'
TP53	NM_001276761	Independent DNA Technologies	Hs.PT.58.39676686	NA*
TERT	NM198253	Independent DNA Technologies	Hs.PT.58.40988589	NA*
ARID2	NM_152641	Independent DNA Technologies	Hs.PT.58.3325252	NA*
EFGR	NM_2001284	Independent DNA Technologies	Hs.PT.58.15419889	NA*
CCND1	NM_053056	Independent DNA Technologies	Hs.PT.56a.4930170	NA*
CCND3	NM_001760	Independent DNA Technologies	Hs.PT.56a.3000774	NA*
ERBB3	NM_001982	Independent DNA Technologies	Hs.PT.58.24466035	NA*
KMT2D	NM_003482	Independent DNA Technologies	Hs.PT.58.4468181	NA*
KMT2C	NM_170606	Independent DNA Technologies	Hs.PT.58.19586130	NA*
TET1	NM_030625	Independent DNA Technologies	Hs.PT.58.20411575	NA*
TET2	NM_001127208	Independent DNA Technologies	Hs.PT.58.26892089	NA*
TET3	NM_144993	Independent DNA Technologies	Hs.PT.58.4763348	NA*

*Sequences of the Independent DNA Technologies inventoried qPCR assays are made available upon purchase.

Protocols

Thawing the Cells

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.

   OCUG-1 cells are thawed and expanded in OCUG-1 Expansion Medium comprising of DMEM (DF-042-B) containing 10% FBS (ES-009-B) and sodium pyruvate (S8636) with optional Penicillin/Streptomycin (P4333).
2. Remove the vial of frozen 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 OCUG-1 Expansion Medium (medium composition in Step 1) to the 15 mL conical tube.

   Important: The expansion medium should be pre-warmed to 37 °C. 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 150 x *g* for 3 minutes to pellet the cells.

   Important: Faster and/or longer centrifugation may result in decreased viability.
8. Decant as much of the supernatant as possible. Steps 5-8 are necessary to remove residual cryopreservative (CryoStor® Cell Cryopreservation Media, C2874).
9. Resuspend the cells in 1-3 mL of OCUG-1 Expansion Medium and determine the cell number and viability.
10. Transfer the cell mixture to a tissue culture flask.

    Important: If the number of the viable cells is 1.5 million or below, use a T75 flask. If the number is above 1.5 million, use a T175 flask. Adjust the volume of the expansion medium accordingly.
11. Incubate the cells at 37 °C in a humidified incubator with 5% CO_2.

Subculturing the Cells

The volumes of the reagents described below are suitable for T75 flasks and should be adjusted for T175 flasks.

1. OCUG-1 cells can be passaged at ~80-85% confluency.
2. Carefully remove the medium from the T75 tissue culture flask containing the 80-85% confluent layer of cells.
3. Rinse the flask with 7-10 mL 1X sterile PBS (TMS-012-A). Aspirate after the rinse. Repeat this wash step one more time.
4. Apply 2 mL of pre-warmed Accutase® (A6964) and incubate in a 37 °C incubator for 3-4 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 4-5 mL of OCUG-1 Expansion Medium to the flask.
7. Gently rotate the flask to mix the cell suspension. Transfer the dissociated cells to a 50 mL conical tube.
8. Centrifuge the tube at 150 x *g* for 3 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 OCUG-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. Recommended density is 1-1.5 million cells for T75 flasks, and 2-3 million cells for T175 flasks.

Cryopreservation of the Cells

The OCUG-1 cells may be frozen in CryoStor® Cell Cryopreservation Media (C2874) using a Nalgene® slow freeze Mr. Frosty® container.

References

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5. Rothenberg SM, Mohapatra G, Rivera MN, Winokur D, Greninger P, Nitta M, Sadow PM, Sooriyakumar G, Brannigan BW, Ulman MJ, Perera RM, Wang R, Tam A, Ma XJ, Erlander M, Sgroi DC, Rocco JW, Lingen MW, Cohen EE, Louis DN, Settleman J, Haber DA. A genome-wide screen for microdeletions reveals disruption of polarity complex genes in diverse human cancers. Cancer Res. 2010 Mar 15;70(6):2158-64.
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