

SK-MEL-1 growing culture | 330424**General information**

Description This cell line was established in 1966 by F. Oettgen and associates using cells from the thoracic duct of a patient. Pigment granules relating both to synthesis and to phagocytosis are present. According to our sequencing, WB and PCR results this cell line carries a BRAF V600E mutation. Cells are N-Ras wildtype.

Organism Human

Tissue Skin

Disease Melanoma

Metastatic site Thoracic lymph duct

Synonyms SK-Mel-1, SK Mel 1, SK-Mel 1, SK-Mel1, SKMEL-1, SkMEL-1, SKMEL1, SK 1

Characteristics

Age 29 years

Gender Male

Ethnicity Caucasian

Morphology Spherical

Growth properties Suspension

Identifiers / Biosafety / Citation

Citation SK-MEL-1 (Cytion catalog number 300424)

Biosafety level 1

Expression / Mutation

Antigen expression Blood Type A, Rh+. Antibody to this line was detected in 63% of patients with malignant melanoma and in 10% of patients with other diseases.

Isoenzymes PGM3, 1, PGM1, 1, ES-D, 1, AK-1, 1, GLO-1, 1-2, G6PD, B,

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Tumorigenic	Yes, in nude mice. Forms pigmented malignant melanomas. Also forms tumors in the cheek pouch of cortisone treated hamsters
Products	Melanin
Mutational profile	V600E type BRAF Mutation was determined by DNA based methods (sequencing, RT-PCR) and protein based methods (Western Blot)

Handling

Culture Medium	EMEM
Medium supplements	10% FBS, w: 2 mM L-Glutamine, w: 1.5 g/L NaHCO ₃ , w: EBSS, w: 1 mM Sodium pyruvate, w: NEAA
Passaging solution	The utilization of a passaging solution is not necessary when passaging cells that are cultured in suspension. The appropriate procedure is to dilute the cells in accordance with the indicated guidelines.
Subculturing	Cultures can be subcultured by addition or replacement of fresh medium. Establish new cultures at 1 x 10 ⁵ viable cells/ml. Maintain at between 1 x 10 ⁵ to 1 x 10 ⁶ cells/ml.
Split ratio	A ratio of 1:2 to 1:4 is recommended
Seeding density	1 x 10 ⁵ cells/mL
Fluid renewal	2 to 3 times per week
Freeze medium	CM-1 (Cytion catalog number 800100) or CM-ACF (Cytion catalog number 806100)

Handling of cryopreserved cultures	The cells come deep-frozen shipped on dry ice. Please make sure that the vial is still frozen. If immediate culturing is not intended, the cryovial must be stored below -150 degree Celsius after arrival. If immediate culturing is intended, please follow the below instructions: Quickly thaw by rapid agitation in a 37 degree Celsius water bath within 40-60 seconds. The water bath should have clean water containing an antimicrobial agent. As soon as the sample has thawed, remove the cryovial from the water bath. A small ice clump should still remain and the vial should still be cold. From now on, all operations should be carried out under aseptic conditions. Transfer the cryovial to a sterile flow cabinet and wipe with 70% alcohol. Carefully open the vial and transfer the cell suspension into a 15 ml centrifuge tube containing 8 ml of culture medium (room temperature). Resuspend the cells carefully. Centrifuge at 300 x g for 3 min and discard the supernatant. The centrifugation step may be omitted, but in this case the remains of the freeze medium have to be removed 24 hours later. Resuspend the cells carefully in 10 ml fresh cell culture medium and transfer them into two T25 cell culture flasks. All further steps are described in the subculture section.
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Handling of proliferating cultures

One or two cell culture flasks come filled with cell culture medium. Collect the entire medium in 1 or 2 x 50 ml centrifuge tubes, respectively. Carefully add 5 ml of cell culture medium to each T25 cell culture flask. Control the cell morphology and confluency under the microscope. Incubate at 37 degree Celsius for a minimum of 24 hours. Spin down the collected medium at 300 x g for 3 minutes to collect the cells which may have detached during transit. If a cell pellet is visible, resuspend the cells in 5 ml of cell culture medium and transfer to a T25 cell culture flask. Incubate at 37 degree Celsius for a minimum of 24 hours.

Quality control / Genetic profile / HLA

Sterility

Mycoplasma contaminations are ruled out through PCR-based and luminescence-based mycoplasma assays. The absence of bacterial, fungal or yeast contamination is controlled through daily visual cell monitoring.

STR profile

Amelogenin: x,Y
CSF1PO: 12,13
D13S317: 11
D16S539: 11,12
D5S818: 12,13
D7S820: 12
TH01: 6
TPOX: 11
vWA: 16,17
D3S1358: 14,16
D21S11: 29,32.2
D18S51: 13,16
Penta E: 7,21
Penta D: 11,13
D8S1179: 13,16
FGA: 18,20

HLA alleles

A*: 26:01:01
B*: 35:01:01, 38:01:01
C*: 04:01:01, 12:03:01
DRB1*: 04:02:01
DQA1*: 03:01:01
DQB1*: 03:02:01
DPB1*: 04:01:01
E: 01:01:01, 01:03:01