

Code:	0287
Cell Line:	Malme-3M
Species:	Homo sapiens
	Vulgar Name: Human
Tissue:	Malignant Melanoma; Derived From Metastatic Site: Lung
Morphology:	Mixed
Disease:	Malignant Melanoma
Growth Properties:	Mixed Adherent-Suspension
Sex:	Male
Age Ethnicity:	43 YEARS; CAUCASIAN
Derivation:	This is one of an extensive series of human tumor lines isolated and characterized by J. Fogh. This melanoma cell line was isolated from the same patient as Malme-3, a normal skin fibroblast.
Applications:	This melanoma cell line was isolated from the same patient as Malme-3, a normal skin fibroblast. The two cell lines provide tumor and normal counterparts for comparative in vitro melanoma studies.
DNA Profile:	Amelogenin: X,Y CSF1PO: 12 D13S317: 8,13 D16S539: 9,12 D5S818: 11 D7S820: 9,12 TH01: 8 TPOX: 8,9 vWA: 15,16
Tumor Formation:	Yes, in nude mice; forms pigmented malignant melanoma
Products:	Antigen expression: HLA A2, Aw30, B13, B40(+/-), DRw7
Biosafety:	1
Culture Medium:	Iscove's Modified Dulbecco's Medium (IMDM) contains 4 mM L-glutamine, 4500 mg/L glucose, and 1500 mg/L sodium bicarbonate. Fetal bovine serum to a final concentration of 20%.
Subculturing:	Volumes used in this protocol are for 75 cm ² flask; proportionally reduce or increase amount of dissociation medium for culture vessels of other sizes. Remove and discard culture medium.

Briefly rinse the cell layer with PBS without calcium and magnesium to remove all traces of serum that contains trypsin inhibitor. Add 2.0 to 3.0 mL of Trypsin-EDTA solution to flask and observe cells under an inverted microscope until cell layer is dispersed (usually within 5 to 15 minutes). Note: To avoid clumping do not agitate the cells by hitting or shaking the flask while waiting for the cells to detach. Cells that are difficult to detach may be placed at 37°C to facilitate dispersal. Add 6.0 to 8.0 mL of complete growth medium and aspirate cells by gently pipetting. Add appropriate aliquots of the cell suspension to new culture vessels. Incubate cultures at 37°C NOTE: For more information on enzymatic dissociation and subculturing of cell lines consult Chapter 12 in Culture of Animal Cells, a manual of Basic Technique by R. Ian Freshney, 6th edition, published by Alan R. Liss, N.Y., 2010.

Medium Renewal: 2 to 3 times per week

Subcultivation ratio: 1:2 to 1:4 is recommended

Culture Conditions:	Atmosphere: air, 95%; carbon dioxide (CO ₂), 5% Temperature: 37°C
Cryopreservation:	95% FBS + 5% DMSO (Dimethyl sulfoxide)
Thawing Frozen Cells:	<p>SAFETY PRECAUTION: Is highly recommend that protective gloves and clothing always be used and a full face mask always be worn when handling frozen vials. It is important to note that some vials leak when submersed in liquid nitrogen and will slowly fill with liquid nitrogen. Upon thawing, the conversion of the liquid nitrogen back to its gas phase may result in the vessel exploding or blowing off its cap with dangerous force creating flying debris.</p> <ol style="list-style-type: none"> 1. Thaw the vial by gentle agitation in a 37°C water bath. To reduce the possibility of contamination, keep the Oring and cap out of the water. Thawing should be rapid (approximately 2 minutes). 2. Remove the vial from the water bath as soon as the contents are thawed, and decontaminate by dipping in or spraying with 70% ethanol. All of the operations from this point on should be carried out under strict aseptic conditions. 3. For cells that are sensitive to DMSO is recommended that the cryoprotective agent be removed immediately. Transfer the vial contents to a centrifuge tube containing 9.0 mL complete culture medium and spin at approximately 125 x g for 5 to 7 minutes. 4. Discard the supernatant and Resuspend cell pellet with the

recommended complete medium (see the specific batch information for the culture recommended dilution ratio). 5. Incubate the culture in a appropriate atmosphere and temperature (see "Culture Conditions" for this cell line). NOTE: It is important to avoid excessive alkalinity of the medium during recovery of the cells. It is suggested that, prior to the addition of the vial contents, the culture vessel containing the growth medium be placed into the incubator for at least 15 minutes to allow the medium to reach its normal pH (7.0 to 7.6).

References:

Human tumor cells in vitro. New York: Plenum Press; 1975. Fogh J, et al. Absence of HeLa cell contamination in 169 cell. Pollack MS, et al. HLA-A, B, C and DR alloantigen expression on forty-six cultured human tumor cell lines. J. Natl. Cancer Inst. 66: 1003-1012, 1981. Fogh J, et al. One hundred and twenty-seven cultured human tumor cell lines producing tumors in nude mice. J. Natl. Cancer Inst. 59: 221-226, 1977. lines derived from human tumors. J. Natl. Cancer Inst. 58: 209-214, 1977.

Depositors:

LETÍCIA VERAS COSTA LOTUFO - Universidade Federal do Ceará

ATCC:

HTB-64