Data Sheet

BCRJ Code:	0325
Cell Line:	Daoy
Species:	Homo sapiens
Vulgar Name:	Human
Tissue:	Brain/Cerebellum
Morphology:	Polygonal
Disease:	Desmoplastic Cerebellar Medulloblastoma
Growth Properties:	Adherent
Sex:	Male
Age/Ethinicity:	4 Year / Caucasian
Derivation:	The Daoy cell line was established in 1985 by P. F Jacobsen of the Royal Perth Hospital in Western Australia. The line was derived from biopsy material taken from a tumor in the posterior fossa of a 4 year old boy.
Applications:	This cell line is a suitable transfection host.
Tumor Formation::	Yes, in nude mice (The cells form serially transplantable intercranial and subcutaneous tumors.)
Biosafety:	1
Addtional Info:	Although the original tumor had characteristics of both neuronal and glial differentiation, these were not retained by the cell line. Treatment of the cells with dibutyryl cyclic amp (cAMP) does not induce expression of those characteristics as measured by staining for S100 (S-100) protein and glial fibrillary acidic proteins (GFAP).

Culture Medium:	Dulbecco's Modified Eagle's Medium (DMEM) with 2 mM L-glutamine, 1.0 g/L glucose and 10% of fetal bovine serum.
Subculturing:	Volumes are given for a 75 cm2 flask. Increase or decrease the amount of dissociation medium needed proportionally 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.
Subculturing Medium Renewal:	2 to 3 times per week
Subculturing Subcultivation Ratio:	1:4 to 1:6 is recommended
Culture Conditions:	Atmosphere: air, 95%; carbon dioxide (CO2), 5% Temperature: 37°C
Cryopreservation:	95% FBS + 5% DMSO (Dimethyl sulfoxide)

Thawing Frozen Cells:	SAFETY PRECAUTION: It is strongly recommended to always wear protective gloves, clothing, and a full-face mask when handling frozen vials. Some vials may leak when submerged in liquid nitrogen, allowing nitrogen to slowly enter the vial. Upon thawing, the conversion of liquid nitrogen back to its gas phase may cause the vial to explode or eject its cap with significant force, creating flying debris. 1. Thaw the vial by gently agitating it in a 37°C water bath. To minimize contamination, keep the O-ring and cap out of the water. Thawing should be rapid (approximately 2 minutes). 2. Remove the vial from the water bath as soon as its contents are thawed and decontaminate it by dipping in or spraying with 70% ethanol. From this point, all operations must be performed under strict aseptic conditions. 3. For cells sensitive to DMSO, it is recommended to remove the cryoprotective agent immediately. Transfer the vial contents to a centrifuge tube containing 9.0 mL of complete culture medium and centrifuge at approximately 125 × g for 5 to 7 minutes. 4. Discard the supernatant and resuspend the cell pellet in the recommended complete medium (see specific batch information for the appropriate dilution ratio). 5. Incubate the culture under appropriate atmospheric and temperature conditions (see "Culture Conditions" for this cell line). NOTE: It is important to avoid excessive alkalinity of the medium during cell recovery. To minimize this risk, it is recommended to place the culture vessel containing the growth medium in the incubator for at least 15 minutes before adding the vial contents. This allows the medium to stabilize at its normal pH (7.0 to 7.6).
References:	He XM, et al. Expression of O6-methylguanine-DNA methyltransferase in six human medulloblastoma cell lines. Cancer Res. 52: 1144-1148, 1992. PubMed: 1737373 Jacobsen PF, et al. Establishment of a human medulloblastoma cell line and its heterotransplantation into nude mice. J. Neuropathol. Exp. Neurol. 44: 472-485, 1985. PubMed: 2993532 Rostomily RC, et al. Expression of neurogenic basic helix-loop-helix genes in primitive neuroectodermal tumors. Cancer Res. 57: 3526-3531, 1997. PubMed: 9270024 The cells form serially transplantable intercranial and subcutaneous tumors.
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