Data Sheet

BCRJ Code:	0420
Cell Line:	LN-18
Species:	Homo sapiens
Vulgar Name:	Human
Tissue:	Brain; Cerebrum; Right temporal lobe
Cell Type:	Epithelial
Morphology:	Epithelial
Disease:	Glioblastoma
Growth Properties:	Adherent
Sex:	Male
Age/Ethinicity:	65 years / White
Derivation:	The LN-18 cell line was established in 1976 from cells taken from a patient with a right temporal lobe glioma.
Applications:	This cell line is used in studies on apoptosis; 3D cell culture; Neuroscience
Products:	Oncogene: p53+ (mutated, TGT (Cys) --> TCT (Ser) mutation at codon 238); PTEN+ (wild-type); p16- (deleted); p14ARF- (deleted) Antigen expression: HLA A2, A9, B5, BW35, DRW3 Genes expressed: fibronectin
Biosafety:	1

Addtional Info:	The cells are negative for glial fibrillary acidic proteins and S100 (S-100) protein. The cells exhibit mutated p53 (TP53) and possible homozygous deletions in the p16 and p14ARF tumor suppressor genes. They have a wild-type PTEN gene. Stimulation of the cells with Fas ligand lead to apoptotic cell death within 16 hours. The cells were also killed by puromycin in a dose dependent manner. Bcl-2 protects these cells from Fas ligand-induced cell death but was shown to have only a small protective effect on puromycin-induced apoptosis. Information received from the depositor indicates the maximum number of serial subcultivations since origin is approximately passage 550. Current lots are available at approximately passage 539.
Culture Medium:	Dulbecco's Modified Eagle's Medium and fetal bovine serum to a final concentration of 5%.
Subculturing:	Volumes used in this protocol are for 75 cm2 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 0.25% (w/v) Trypsin-0.53% (w/v) EDTA solution 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
Subculturing Subcultivation Ratio:	1:4 to 1:6
Culture Conditions:	Atmosphere: air, 95%; carbon dioxide (CO2), 5%
Cryopreservation:	95% FBS + 5% DMSO (Dimethyl sulfoxide)

Thawing Frozen Cells:	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. 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:	Diserens AC, et al. Characterization of an established human malignant glioma cell line: LN-18. Acta Neuropathol. 53: 21-28, 1981. PubMed: 7211194 Ishii N, et al. Frequent co-alterations of TP53, p16/CDKN2A, p14ARF, PTEN tumor suppressor genes in human glioma cell lines. Brain Pathol. 9: 469-479, 1999. PubMed: 10416987 Schlapbach R, Fontana A. Differential activity of bcl-2 and ICE enzyme family protease inhibitors on Fas and puromycin-induced apoptosis of glioma cells.. Biochim. Biophys. Acta 1359: 174-180, 1997. PubMed: 9409814 Flaman JM, et al. A simple p53 functional assay for screening cell lines, blood, and tumors. Proc. Natl. Acad. Sci. USA 92: 3963-3967, 1995. PubMed: 7732013
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