

Data Sheet

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BCRJ Code:	0101
Cell Line:	HEp-2
Species:	Homo sapiens
Vulgar Name:	Human
Tissue:	Hela Contaminant
Morphology:	Epithelial
Disease:	Carcinoma
Growth Properties:	Adherent
Age/Ethnicity:	56 Year /
Virus Susceptibility::	Human adenovirus 3 Human poliovirus 1 Vesicular stomatitis virus
Biosafety:	2
Additional Info:	Cells of this line contain HeLa marker chromosomes, and were derived via HeLa contamination. This line was originally thought to be derived from an epidermoid carcinoma of the larynx, but was subsequently found, based on isoenzyme analysis, HeLa marker chromosomes, and DNA fingerprinting, to have been established via HeLa cell contamination. The cells are positive for keratin by immunoperoxidase staining. ATCC confirmed this cell line is positive for the presence of human papilloma viral DNA sequences via PCR.
Culture Medium:	Dulbecco's Modified Eagle's Medium (DMEM) with 1% non-essential amino acids, 2 mM L-glutamine, 1.0 g/L glucose and 10% of fetal bovine serum.

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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. T-75 flasks are recommended for subculturing this product. 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 1:10

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 highly recommended 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 submerged 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 vial 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 it 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 an 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:

Moore AE, et al. Culture characteristics of four permanent lines of human cancer cells. *Cancer Res.* 15: 598-602, 1955. PubMed: 13261081 Chen TR. Re-evaluation of HeLa, HeLa S3, and HEp-2 karyotypes. *Cytogenet. Cell Genet.* 48: 19-24, 1988. PubMed: 3180844 Toolan HW. Transplantable human neoplasms maintained in cortisone-treated laboratory animals: H.S. No. 1; H.Ep. No. 1; H.Ep. No. 2; H.Ep. No. 3; and H.Emb.Rh. No. 1. *Cancer Res.* 14: 660-666, 1954. PubMed: 13209540 Black FL, et al. Propagation of measles virus in a strain of human epidermoid cancer cells (Hep-2). *Proc. Soc. Exp. Biol. Med.* 93: 107-108, 1956. PubMed: 13370591 . . Tex. Rep. Biol. Med. 15: 588, 1957. Moore AE. Tumorigenic activity of cultures. *Ann. N.Y. Acad. Sci.* 76: 497-505, 1958. PubMed: 13627875 St. Geme JW, et al. Characterization of the genetic locus encoding Haemophilus influenzae type b surface fibrils. *J. Bacteriol.* 178: 6281-6287, 1996. PubMed: 8892830 Gromeier M, et al. Internal ribosomal entry site substitution eliminates neurovirulence in intergeneric poliovirus recombinants. *Proc. Natl. Acad. Sci. USA* 93: 2370-2375, 1996. PubMed: 8637880 Roller RJ, et al. Structure and function in the herpes simplex virus 1 RNA-binding protein US11: mapping of the domain required for ribosomal and nucleolar association and RNA binding in vitro. *J. Virol.* 70: 2842-2851, 1996. PubMed: 8627758 Herold BC, et al. Differences in the susceptibility of herpes simplex virus types 1 and 2 to modified heparin compounds suggest serotype differences in viral entry. *J. Virol.* 70: 3461-3469, 1996. PubMed: 8648678 Chang YE, et al. Properties of the protein encoded by the UL32 open reading frame of herpes simplex virus 1. *J. Virol.* 70: 3938-3946, 1996. PubMed: 8648731 Carter KL, et al. Characterization of the products of the UL43 gene of herpes simplex virus 1: potential implications for regulation of gene expression by antisense transcription. *J. Virol.* 70: 7663-7668, 1996. PubMed: 8892886 Carter KL, Roizman B. The promoter and transcriptional unit of a novel herpes simplex virus 1 alpha gene are contained in, and encode a protein in frame with, the open reading frame of the alpha22 gene. *J. Virol.* 70: 172-178, 1996. PubMed: 8523523 Jamaluddin M, et al. Inducible translational regulation of the NF-IL6 transcription factor by respiratory syncytial virus infection in pulmonary epithelial cells. *J. Virol.* 70: 1554-1563, 1996. PubMed: 8627674

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CCL-23