Cell Lines

Murine Tumor (Catalog No. JHU-1): This cell line was developed to circumvent the issue of negatively selecting nonessential tumor markers in cancer vaccine experiments. Primary lung epithelial cells from C57BL/6 mice were immortalized with HPV-16 E6 and E7 and then transformed with the activated *ras* oncogene. This transformation process mimics the natural sequence in the pathogenesis of cervical cancer in which HPV-16 plays a critical role.

Lin KY et al. Treatment of established tumors with a novel vaccine that enhances major histocompatibility class II presentation of tumor antigen. Cancer Res. 56: 21-26, 1996; PubMed: 8548765.

Rat Prostate Tumor Cell Lines

The cell line **R-3327-G (Catalog No. JHU-3)** was derived from the parental R3327 tumor discovered by W.F. Dunning in a 22-month-old inbred male rat. After many years of serial passages the H subline was characterized as a slow-growing heterogeneous tumor composed of both androgen-dependent and androgen independent tumor cells. While passaging the H subline, random tumor progression gave rise to fast-growing anaplastic tumors (AT-1, AT-2, and AT-3). The AT-1 subline was further passed and gave rise the MAT-Lu and MAT-LyLu.

The AT-1, AT-2, and AT-3 sublines can be used to study the effects of various types of cancer therapies on prostate cells. They also provide a model for studying prostate malignancies in vitro and in vivo.

Rat Prostate R3327-AT-1 Tumor (Catalog No. JHU-29): AT-1 was the first anaplastic tumor to arise from the H subline. Following subcutaneous transplantation in syngeneic male rats, AT-1 forms solid sheets of malignant cells with no indication of glandular function. The cells are androgen independent, have a doubling time of approximately 2.5 days, and exhibit low metastatic ability (<5%).

Rat Prostate R3327-AT-2.1 Tumor (Catalog No. JHU-30): AT-2 was the second anaplastic tumor to arise from the H subline. Following subcutaneous transplantation in syngeneic male rats, AT-2 forms solid sheets of malignant cells with no indication of glandular function. The cells are androgen independent, have a doubling time of approximately 2.5 days, and exhibit low to moderate metastatic ability (<20%).

Rat Prostate R3327-AT-3.1 Tumor (Catalog No. JHU-31): By growing the H subline in castrated male rats, the androgen-independent HI-S subline was established in 1978. Continuous serial passage of HI-S in castrated male rats gave rise to the less differentiated fast-growing subline, HI-F. AT-3.1 is a completely anaplastic tumor subline which arose within a particular passage of HI-F in 1982. Following subcutaneous transplantation in syngeneic male rats, AT-3 forms solid sheets of malignant cells with no indication of glandular function. The cells are androgen-independent, have a doubling time of 1.5-1.8 days, and exhibit a high rate of metastasis to the lung and lymph nodes (>75%).

After continuous serial passage, AT-1 gave rise to two distinct, metastatic anaplastic sublines which are valuable models for studying prostate malignancies both in vitro and in vivo.

Rat Prostate MAT-Lu (Catalog No. JHU-4): Produces lung metastases almost exclusively.
Rat Prostate MAT-LyLu (Catalog No. JHU-5): Produces metastases to both the lymph nodes and lung.

Isaacs JT et al. Establishment and characterization of seven Dunning rat prostatic cancer cell lines and their use in developing methods for predictingmetastatic abilities of prostatic cancers. Prostate 9(3): 261-281, 1986; PubMed 87041052.
Isaacs JT. Genetic instability coupled to clonal selection as a mechanism for tumor progression in the Dunning R-3327 rat prostatic adenocarcinoma system. Cancer Res. 42(6): 2353-2371, 1982; PubMed 7074614.
Isaacs JT et al. The characterization of a newly identified, highly metastatic variety of Dunning R 3327 rat prostatic adenocarcinoma system: the MAT LyLu tumor. Invest. Urol. 19(1):20-23, 1981; PubMed 7251319.
Claflin AJ et al. The Dunning R3327 prostate adenocarcinoma in the Fischer-Copenhagen F1 rat: a useful model for immunological studies. Oncology 34(3): 105-109, 1977; PubMed 917439.

Block NL et al. Chemotherapy of the transplantable adenocarcinoma (R-3327) of the Copenhagen rat. Oncology 34(3): 110-113, 1977; PubMed 917440.
Dunning WF. Prostate cancer in the rat. Monographs of the National Cancer Institute 12: 351-369, 1963.

Rat 1A Fibroblast (Catalog No. JHU-25): This rat cell line was derived from spontaneously immortalized rat embryo fibroblasts isolated from day 14 Fisher rat embryos and can be used as a control for JHU-26 (below).

Small MB et al. Neoplastic transformation by the human gene N-myc. Mol. Cell. Biol. 7: 1638-1645, 1987; PubMed 3299052.

Rat 1A-Myc Fibroblast (Catalog No. JHU-26): Rat 1A-myc fibroblasts were generated by cotransfection of MLV long terminal repeat-driven genomic c-*myc* sequence (pMLVmyc) with a neomycin-resistance marker plasmid (pSV2neo) into rat-1A fibroblasts (JHU-25). Cells that overexpress Myc were selected with the antibiotic G418. This cell line may be useful in the study of cell division or cell death.

Hoang AT et al. Participation of cyclin A in Myc-induced apoptosis. Proc. Natl. Acad. Sci. USA 91: 6875-6879, 1994; PubMed 8041712.

Human Primary Effusion Lymphoma JSC-1 (Catalog No. JHU-32): This primary effusion lymphoma (PEL) cell line was established from lymphomatous peritoneal effusion tumor cells that were anaplastic, large, and hematopoietic in appearance. The resultant cell line has a similar morphology and phenotype with strong CD45 and CD71, partial CD20, and lambda light chain restriction by flow cytometry. JSC-1 is positive for Kaposi's sarcoma herpesvirus (KSHV) and type 1 Epstein-Barr virus and yields virions that are highly infectious to primary endothelial cell cultures. It exhibits higher basal and induced expression of KSHV lytic cycle gene products than do other established PEL cell lines, making it an easily controlled and managed system for studying primary infection in vitro and the role of KSHV in pathogenesis of certain disease states.

Cannon JS et al. A new primary effusion lymphoma-derived cell line yields a highly infectious Kaposi's sarcoma herpesvirus-containing upernatant. J. Virol. 74(21): 10187-93 2000 PubMed 11024147.

Human Bronchial Epithelial Cystic Fibrosis Cell Lines

IB3-1 (Catalog No. JHU-52): IB3-1 cells were derived from a cystic fibrosis patient with compound heterozygous mutations (ΔF_{508}/W1282X) in the gene coding for the cystic fibrosis transmembrane conductance regulator (CFTR). Cells were isolated from the bronchial epithelia and immortalized by viral transformation. These cells retain characteristics of epithelial cells and are deficient in protein kinase A-mediated activation of chloride conductance, which is diagnostic of the CF genetic defect. IB3-1 cells are an appropriate substrate for studies of the mutant CFTR protein and its interaction with the Cl^- channel. IB3-1 cells can also provide important information concerning the effects of alternate mutations or heterozygote states of ΔF_{508} on defective Cl^- transport.

S9 clone (Catalog No. JHU-53): IB3-1 cells were transfected with a cDNA encoding a CFTR protein to generate the S9 clone. Expression of CFTR is driven by the adeno-associated virus type 2 inverted terminal repeat. Stimulation of Cl- efflux by forskolin and 3-isobutyl-1-methylxanthine was not seen in CFTR-mutant IB3-1 cells but was evident in S9 cells demonstrating the functional reconstitution of CFTR in S9 cells.

C38 clone (Catalog No. JHU-54): IB3-1 cells were transfected with a cDNA encoding a CFTR protein to generate the C38 clone. Expression of this functional CFTR, which contains a deletion of nucleotides 123-486 in the amino-terminal region of the cDNA, is driven by the adeno-associated virus type 2 inverted terminal repeat. When compared to the parental IB3-1 cell line, C38 cells exhibit an increased basal rate of Cl^- efflux and an elevated efflux rate upon forkskolin treatment, as expected for normal CFTR function.

Flotte TR et al. Adeno-associated virus vector gene expression occurs in nondividing cells in the absence of vector DNA integration. Am. J. Respir. Cell. Mol. Biol. 11(5): 517-521, 1994; PubMed 7946381.
Flotte TR et al. Expression of the cystic fibrosis transmembrane conductance regulator from a novel adeno-associated virus promoter. J. Biol. Chem. 268(5): 3781-3790, 1993; PubMed 7679117.
Egan M et al. Defective regulation of outwardly rectifying Cl- channels by protein kinase A corrected by insertion of CFTR. Nature 358(6387): 581-584, 1992; PubMed 1380129.
Flotte TR et al. Gene expression from adeno-associated virus vectors in airway epithelial cells. Am. J. Respir. Cell. Mol. Biol. 7(3): 349-356, 1992; PubMed 1325813.
Zeitlin PL et al. A cystic fibrosis bronchial epithelial cell line: immortalization by adeno-12-SV40 infection. Am. J. Respir. Cell. Mol. Biol. 4(4):313-319A, 1991; PubMed 1849726.

p53R Cell Line (Catalog No. JHU-56): The p53R cell line was established by the stable transfection of HS 766T cells (ATCC HTB-134)

with DNA consisting of four tandem copies of the internally symmetric Tp53 consensus DNA binding site located upstream of the SV40 minimal viral promoter and a luciferase reporter gene. The parental cell line, HS 766T, was isolated from a pancreatic carcinoma metastatic to a lymph node and contains the wild-type TP53 gene. The p53R cell line offers an inexpensive, high-throughput assay for Tp53 activation in human cells.

Sohn TA et al. High-throughput measurement of the Tp53 response to anticancer drugs and random compounds using a stably integrated Tp53-responsive luciferase reporter. Carcinogenesis 23(6): 949-957, 2002.

Sohn TA et al. High-throughput drug screening of the DPC4 tumor-suppressor pathway in human pancreatic cancer cells. Ann. Surg. 233(5): 696-703, 2001.

Hybridomas

Anti Qa-1b Hybridomas: B cells from B6-T1a^a mice immunized with peptide (161-179aa) from the unique α-2 domain of the Qa-1^b molecule were fused to the murine B cell myeloma, P3x653, to generate these cloned hybridomas. Qa-1^b is a nonclassical MHC class I (Class Ib) molecule and is the murine counterpart to HLA-E. Each hybridoma produces murine anti-Qa-1^{b}monoclonal antibody of the IgG1k isotype that can be used for ELISA, flow cytometry, western blotting, immunoprecipitation, and immunocytochemistry.

Anti Qa-1b Hybridoma 6A8.6F10.1A6: Catalog No: CRL-2743 (JHU-27)
Anti Qa-1b Hybridoma 4C2.4A7.5H11: Catalog No. CRL-2744 (JHU-28)

Gays F et al. Functional analysis of the molecular factors controlling Qa1-mediated protection of target cells from NK lysis. J. Immunol.16:1601-1610, 2001; PubMed 11160201.

Lo W-F et al. Cytotoxic T cell responses to gram-negative intracellular bacterial pathogens: A role for CD8+ in immunity to Salmonella infection and involvement of Class Ib molecules. J. Immunol.162: 5398-5406, 1999; PubMed 10228017.

Vectors

MoPrP.Xho Expression Vector (Catalog No. JHU-2): This plasmid expression vector was developed to direct the expression of foreign genes in the brains of transgenic mice. The vector was derived from segments of genomic DNA that compose the mouse prion protein gene, specifically from the promoter, 5' intronic, and 3' untranslated sequences. The vector has been used in multiple settings to produce mice that express human genes in brain (both in neurons and glia throughout the nervous system) and muscle.

Thinakaran G et al. Endoproteolysis of presenilin 1 and accumulation of processed derivatives in vivo. Neuron 17:181?190, 1996; PubMed 11337275.

Borchelt DR. A vector for expressing foreign genes in the brains and hearts of transgenic mice. Genet. Anal. 13(6):159-163, 1996; 9117892.

Clones

Gene product: Hypoxia-inducible factor 1 alpha [HIF1 alpha]

pCEP4/HIF-1alpha: Catalog No. MBA-2 (JHU-33)
pBluescriptSK/HIF-1alpha: Catalog No. MBA-3 (JHU-34)
pCEP4HIF-1alphaDN, pCEP/HIF-1alpha deltaNB deltaAB: Catalog No. MBA-7 (JHU-38)
pGalA, pGAL-HIF-1alpha (531-826): Catalog No. MBA-9 (JHU-40)

Jiang BH et al. Transactivation and inhibitory domains of hypoxia-inducible factor 1alpha. J. Biol. Chem. 272: 19253-19260, 1997; PubMed 9235919.

Jiang et al. Dimerization, DNA binding, and transactivation properties of hypoxia-inducible factor 1. J. Biol. Chem. 271: 17771-17778, 1996; PubMed 8663540.

Wang GL et al. Hypoxia-inducible factor 1 is a basic-helix-loop-helix-PAS heterodimer regulated by cellular O_2 tension. Proc. Natl. Acad. Sci. USA 92: 5510-5514, 1995; PubMed 7539918.

Gene product: VEGF promoter

pVEGF-KpnI: Catalog No. MBA-4 (JHU-35)
p11w: Catalog No. MBA-10 (JHU-41)
p11m: Catalog No. MBA-11 (JHU-42)

Forsythe JA et al. Activation of Vascular Endothelial Growth Factor Gene Transcription by Hypoxia-Inducible Factor 1. Mol. Cell. Biol. 16: 4604-4613, 1996; PubMed 8756616.

Gene product: Enolase 1 promoter

p2.1, pAGL2A: Catalog No. MBA-5 (JHU-36)
p2.4, pAGL2D: Catalog No. MBA-6 (JHU-37)

Semenza GL et al. Hypoxia response elements in the aldolase A, enolase 1, and lactate dehydrogenase A gene promoters contain essential binding sites for hypoxia-inducible factor 1. J. Biol. Chem. 271: 32529-32537, 1996; PubMed 8955077.

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Kits

The AdEasy® System

The AdEasy system employs the recombination processes in *E. coli* to produce recombinant adenovirus quickly and efficiently. The gene of interest is cloned into a shuttle vector and then inserted into the adenovirus genome backbone in the *E. coli* cells. The *E. coli* plasmid is used to transfect a packaging cell line such as 293 cells. The system avoids multiple plaque assays associated with traditional adenovirus transfection and is thus a faster, simpler route to gene transfer and expression in mammalian cells.

AdEasy Basic Kit (Catalog No. JHU-23) contains the *E. coli* cells and plasmids necessary to product recombinant virus:

AdEasier™-1 Cells (BJ5183 strain containing pAdEasy-1)
pShuttle in *E. coli* DH10B
pShuttle-CMV in DH10B
pAdTrack in DH10B
pAdTrack-CMV in DH10B

AdEasy Supplement Kit (Catalog No. JHU-24) contains components to test the efficiency of the AdEasy system for making recombinant adenoviruses.

E. coli BJ5183 cells
pAdEasy-1 in *E. coli* DH10B
pAdEasy-2 in DH10B
pAdEasy-1-GFP+ßgal in DH10B
pAdEasy-2-GFP+ßgal in DH10B

He T-C et al. A simplified system for generating recombinant adenoviruses. Proc. Natl. Acad. Sci. USA 95: 2509-2514, 1998.
U.S. Patent 5,922,576 dated July 13, 1999.

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The CMV promoter used in some of the constructs and the polyadenylation site were both from pEGFP-C1 and obtained from Clontech (Palo Alto, CA). The CMV promoter is covered under U.S. Patents 5,168,062 and 5,385,839 and its use is permitted for research purposes only. Any other use of the CMV promoter requires a license from the University of Iowa Research Foundation, 214 Technology Innovation Center, Iowa City, IA 52242. Use of Clontech's Living Colors™ products containing DNA sequences coding for mutant *Aequorea victoria* green fluorescent protein (GFP) variants or proteins thereof requires a license from Aurora Biosciences Corporation under U.S. Patent Nos. 5,625,048, 5,777,049, 6,054,321, and 5,804,387 and other pending U.S. and foreign patent applications. In addition, certain Clontech products are made under U.S. Patent No. 5,804,387 licensed from Stanford University.

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