BCRJ Code:	0041
Cell Line:	ARPE-19
Species:	Homo sapiens
Vulgar Name:	Human
Tissue:	Retinal Pigmented Epithelium; Retina
Cell Type:	Epithelial
Morphology:	Epithelial
Disease:	Normal
Growth Properties:	Adherent
Sex:	Male
Age/Ethinicity:	19 Year /
Derivation:	ARPE-19 is a spontaneously arising retinal pigment epithelia (RPE) cell line derived from the normal eyes of a 19-year-old male who died from head trauma in a motor vehicle accident.
Applications:	This cell line is a suitable transfection host.
DNA Profile:	Amelogenin: X,Y CSF1PO: 11 D13S317: 11,12 D16S539: 9,11 D5S818: 13 D7S820: 9,11 THO1: 6,9.3 TPOX: 9,11 vWA: 16,19
Biosafety:	1

Addtional Info:	These cells form stable monolayers, which exhibit morphological and functional polarity. ARPE-19 expresses the RPE-specific markers CRALBP and RPE-65. The cells exhibit morphological polarization when plated on laminin-coated Transwell-COL filters in medium with a low serum concentration. They form tight-junctions with transepithelial resistance of monolayers reaching a maximum of 50 to 100 ohms/cm2 after 4 weeks of culture.
Culture Medium:	1:1 mixture of Dulbecco's modified Eagle's medium and Ham's F12 medium containing 2.5 mM L-glutamine, 0.5 mM sodium pyruvate and fetal bovine serum to a final concentration of 10%.
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:	Every 2 to 3 days
Subculturing Subcultivation Ratio:	1:3 to 1:5
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:

Dunn KC, et al. ARPE-19, A human retinal pigment epithelial cell line with differentiated properties. Exp. Eye Res. 62: 155-169, 1996. PubMed: 8698076 Maidji E, et al. Accessory human cytomegalovirus glycoprotein US9 in the unique short component of the viral genome promotes cell-to-cell transmission of virus in polarized epithelial cells. J. Virol. 70: 8402-8410, 1996. PubMed: 8970961 Holtkamp GM, et al. Polarized secretion of IL-6 and IL-8 by human retinal pigment epithelial cells. Clin. Exp. Immunol. 112: 34-43, 1998. PubMed: 9566787 Finnemann SC, et al. Phagocytosis of rod outer segments by retinal pigment epithelial cells requires alpha(v)beta5 integrin for binding but not for internalization. Proc. Natl. Acad. Sci. USA 94: 12932-12937, 1997. PubMed: 9371778 Handa JT, et al. The advanced glycation endproduct pentosidine induces the expression of PDGF-B in human retinal pigment epithelial cells. Exp. Eye Res. 66: 411-419, 1998. PubMed: 9593635 Dunn KC, et al. Use of the ARPE-19 cell line as a model of RPE polarity: basolateral secretion of FGF5.. Invest. Ophthalmol. Vis. Sci. 39: 2744-2749, 1998. PubMed: 9856785 Tugizov S, et al. An acidic cluster in the cytosolic domain of human cytomegalovirus glycoprotein B is a signal for endocytosis from the plasma membrane. J. Virol. 73: 8677-8688, 1999. PubMed: 10482621 Orten DJ, et al. Analysis of DNA elements that modulate myosin VIIA expression in humans. Hum. Mutat. 14: 354, 1999. PubMed: 10502787 Rajan PD, et al. Expression of the extraneuronal monoamine transporter in RPE and neural retina. Curr. Eye Res. 20: 195-204, 2000. PubMed: 10694895 Janssen JJ, et al. Retinoic acid delays transcription of human retinal pigment neuroepithelium marker genes in ARPE-19 cells. Neuroreport 11: 1571-1579, 2000. PubMed: 10841379 Udono T, et al. Adrenomedullin in cultured human retinal pigment epithelial cells. Invest. Ophthalmol. Vis. Sci. 41: 1962-1970, 2000. PubMed: 10845623

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