

Code:	0070
Cell Line:	COS-1
Species:	Cercopithecus aethiops Vulgar Name: Monkey; African Green Monkey
Tissue:	Kidney
Morphology:	Fibroblast
Growth Properties:	Adherent
Sex:	Male
Age Ethnicity:	ADULT
Applications:	This is an African green monkey kidney fibroblast-like cell line suitable for transfection by vectors requiring expression of SV40 T antigen. This line contains T antigen, retains complete permissiveness for lytic growth of SV40, supports the replication of ts A209 virus at 40C, and supports the replication of pure populations of SV40 mutants with deletions in the early region.
Virus Susceptibility:	SV40 virus
Products:	T-antigen
Biosafety:	2
Additional info:	The cells contain a single integrated copy of the complete early region of the SV40 genome.
Culture Medium:	Dulbecco's modified Eagle's medium with 4 mM L-glutamine adjusted to contain 1.5 g/L sodium bicarbonate and 4.5 g/L glucose and supplemented with 5 mM HEPES, 90%; fetal bovine serum, 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.

Medium Renewal: 2 to 3 times per week

Subcultivation ratio: 1:4 to 1:8

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 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 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:

Gluzman Y. SV40-transformed simian cells support the replication of early SV40 mutants. *Cell* 23: 175-182, 1981. PubMed: 6260373 Mansky LM. The mutation rate of human immunodeficiency virus type 1 is influenced by the vpr gene. *Virology* 222: 391-400, 1996. PubMed: 8806523 Churchill MJ, et al. The rev-responsive element negatively regulates human immunodeficiency virus type 1 env mRNA expression in primate cells. *J. Virol.* 70: 5786-5790, 1996. PubMed: 8709194 Goodrum FD, et al. Adenovirus early region 4 34-kilodalton protein directs the nuclear localization of the early region 1B 55-kilodalton protein in primate cells. *J. Virol.* 70: 6323-6335, 1996. PubMed: 8709260 Suss-Toby E, et al. Toxoplasma invasion: the parasitophorous vacuole is formed from host cell plasma membrane and pinches off via a fission pore. *Proc. Natl. Acad. Sci. USA* 93: 8413-8418, 1996. PubMed: 8710885 Chang K, Pastan I. Molecular cloning of mesothelin, a differentiation antigen present on mesothelium, mesotheliomas, and ovarian cancers. *Proc. Natl. Acad. Sci. USA* 93: 136-140, 1996. PubMed: 8552591 Lu FM, Lux SE. Constitutively active human notch 1 binds to the transcription factor CBF1 and stimulates transcription through a promoter containing a CBF1-responsive element. *Proc. Natl. Acad. Sci. USA* 93: 5663-5667, 1996. PubMed: 8643633 Bhattacharyya DK, et al. Involvement of arginine 120, glutamate 524, and tyrosine 355 in the binding of arachidonate and 2-phenylpropionic acid inhibitors to the cyclooxygenase active site of ovine prostaglandin endoperoxide H synthase-1. *J. Biol. Chem.* 271: 2179-2184, 1996. PubMed: 8567676 Feng XH, Derynck R. Ligand-independent activation of transforming growth factor (TGF) beta-signaling pathways by heteromeric cytoplasmic domains of TGF-beta receptors. *J. Biol. Chem.* 271: 13123-13129, 1996. PubMed: 8662796 Wang LH, et al. Identification of thromboxane A2 synthase active site residues by molecular modeling-guided site-directed mutagenesis. *J. Biol. Chem.* 271: 19970-19975, 1996. PubMed: 8702713 Almaula N, et al. Mapping the binding site pocket of the serotonin 5-hydroxytryptamine 2A receptor. *J. Biol. Chem.* 271: 14672-14675, 1996. PubMed: 8663249

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ATCC:

CRL-1650