BCRJ Code:	0323
Cell Line:	T-47D
Species:	Homo sapiens
Vulgar Name:	Human
Tissue:	Mammary Gland; Derived From Metastatic Site: Pleural Effusion
Cell Type:	Epithelial
Morphology:	Epithelial
Disease:	Ductal Carcinoma
Growth Properties:	Adherent
Sex:	Female
Age/Ethinicity:	54 Year /
Derivation:	The T-47 line was isolated by I. Keydar from a pleural effusion obtained from a 54 year old female patient with an infiltrating ductal carcinoma of the breast.
Applications:	This cell line is a suitable transfection host.
Biosafety:	1
Addtional Info:	This differentiated epithelial substrain (T-47D) was found to contain cytoplasmic junctions and receptors to 17 beta estradiol, other steroids and calcitonin. The cells express the WNT7B oncogene.
Culture Medium:	RPMI-1640 medium modified to contain 2 mM L-glutamine, 1 mM sodium pyruvate, 4500 mg/L glucose and 10% of fetal bovine serum.

Subculturing:	Volumes are given for a 75 cm2 flask. Increase or decrease the amount of dissociation medium needed proportionally for culture vessels of other sizes. Corning® T-75 flasks (catalog #430641) 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. Population Doubling Time 32 hrs 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:	1:3 to 1:5 is recommended
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:

Judge SM, Chatterton RT Jr.. Progesterone-specific stimulation of triglyceride biosynthesis in a breast cancer cell line (T-47D). Cancer Res. 43: 4407-4412, 1983. PubMed: 6871874 Lamp SJ, et al. Calcitonin induction of a persistent activated state of adenylate cyclase in human breast cancer cells (T-47D). J. Biol. Chem. 256: 12269-12274, 1981. PubMed: 6271778 Sher E, et al. Whole-cell uptake and nuclear localization of 1,25-dihydroxy-cholecalciferol by breast cancer cells (T-47D) in culture. Biochem. J. 200: 315-320, 1981. PubMed: 6896147 Freake HC, et al. 1,25-Dihydroxyvitamin D3 specifically binds to a human breast cancer cell line (T-47D) and stimulates growth. Biochem. Biophys. Res. Commun. 101: 1131-1138, 1981. PubMed: 6272774 Faust JB, Meeker TC. Amplification and expression of the bcl-1 gene in human solid tumor cell lines. Cancer Res. 52: 2460-2463, 1992. PubMed: 1568216 Huguet EL, et al. Differential expression of human Wnt genes 2, 3, 4, and 7B in human breast cell lines and normal and disease states of human breast tissue. Cancer Res. 54: 2615-2621, 1994. PubMed: 8168088 Bellet D, et al. Malignant transformation of nontrophoblastic cells is associated with the expression of chorionic gonadotropin beta genes normally transcribed in trophoblastic cells. Cancer Res. 57: 516-523, 1997. PubMed: 9012484 Hoppe HC, et al. Identification of phosphatidylinositol mannoside as a mycobacterial adhesin mediating both direct and opsonic binding to nonphagocytic mammalian cells. Infect. Immun. 65: 3896-3905, 1997. PubMed: 9284169 Burfeind P, et al. Antisense RNA to the type I insulin-like growth factor receptor suppresses tumor growth and prevents invasion by rat prostate cancer cells in vivo. Proc. Natl. Acad. Sci. USA 93: 7263-7268, 1996. PubMed: 8692980 Keydar I, et al. Establishment and characterization of a cell line of human breast carcinoma origin. Eur. J. Cancer 15: 659-670, 1979. PubMed: 228940

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