BT-474

300131

GENERAL INFORMATION

Description

BT-474 is a human breast cancer cell line, derived from the ductal carcinoma of a 60-year-old female. This cell line is estrogen and progesterone receptor positive, making it a valuable model for studying hormone-responsive breast cancers. BT-474 cells are also characterized by the overexpression of HER2/neu (human epidermal growth factor receptor 2), a protein that is amplified and plays a critical role in the pathogenesis and progression of certain aggressive types of breast cancer.
The BT-474 cell line is extensively used in oncological research to study the molecular mechanisms of breast cancer proliferation and to test therapeutic strategies targeting hormone receptors and the HER2 pathway. These cells are particularly useful for examining the efficacy of HER2-targeted therapies, such as trastuzumab (Herceptin), and for exploring mechanisms of resistance to these treatments. The cell line has also contributed to advancements in understanding how hormonal manipulations affect cancer cell growth and survival, providing insights into potential treatment approaches for hormone-dependent tumors.

Tissue	Breast, mammary gland
Disease	Invasive ductal carcinoma
Metastatic site	Ductal

Synonyms

- Bt-474
- BT474

CLASSIFICATION

Organism	Human
Sex	Female
Growth properties	Adherent
Biosafety level	1

CHARACTERISTICS

Morphology	Epithelial-like
Tumourigenic	Yes, in nude mice
MSI status	Stable (MSS)

Karyotype

Mode = 55, range = 50 to 112, bimodal shift 58 - 59 and 100 in later passages with 3 marker chromosomes

Doubling time	60 to 80 hours

IDENTIFIERS

Citation

BT-474 (Cytion catalog number 300131)

NCBI TaxID	9606

Cytion GmbH
Nikola-Tesla-Str. 3
69124 Heidelberg
Tel: +49 (0)6221 405780
info@cytion.com
cytion.com

Registered office Heidelberg
VAT ID: DE283292917
Local court Mannheim - HRB 714394
CEO / Geschäftsführer: Jonathan Steubing

Banking information
Sparkasse Heidelberg
IBAN: DE35 6725 0020 0009 2076 86
BIC: SOLADES1HDB

Cellosaurus	CVCL_0179

EXPRESSION AND SUSCEPTIBILITY

Receptors expressed

HER-2/NEU+, ER+, PR+

Isoenzymes

G6PD, B, PGM3, 1, PGM1, 1, ES-D, 1, Me-2, 0, AK-1, 1, GLO-1, 1, Phenotype Frequency Product: 0.0426

Mutational profile

TP53 mut

Virus susceptibility

Mouse mammary tumor virus (RIII-MuMTV)

HANDLING AND CULTURE

Culture medium

DMEM:Ham's F12 (1:1), w: 3.1 g/L Glucose, w: 2.5 mM L-Glutamine, w: 15 mM HEPES, w: 0.5 mM Sodium pyruvate, w: 1.2 g/L NaHCO3 (Cytion article number 820400a)

Incubation atmosphere	37°C, 5% CO_2, humidified atmosphere.

Supplements

Supplement the medium with 10% FBS, 10 microgram/mL Insulin

Subculturing

Remove the old medium from the adherent cells and wash them with PBS that lacks calcium and magnesium. For T25 flasks, use 3-5 ml of PBS, and for T75 flasks, use 5-10 ml. Then, cover the cells completely with Accutase, using 1-2 ml for T25 flasks and 2.5 ml for T75 flasks. Let the cells incubate at room temperature for 8-10 minutes to detach them. After incubation, gently mix the cells with 10 ml of medium to resuspend them, then centrifuge at 300xg for 3 minutes. Discard the supernatant, resuspend the cells in fresh medium, and transfer them into new flasks that already contain fresh medium.

Seeding density	2 x 10^4 cells/cm^2 will yield in a mostly confluent layer in about 4 days
Fluid renewal	2 to 3 times per week
Post-thaw recovery	Almost 100% recovered cells at >90% viability

Thawing and culturing

Confirm that the vial remains deeply frozen upon delivery, as cells are shipped on dry ice to maintain optimal temperatures during transit. Upon receipt, either store the cryovial immediately at temperatures below -150°C to ensure the preservation of cellular integrity, or proceed to step 3 if immediate culturing is required.
For immediate culturing, swiftly thaw the vial by immersing it in a 37°C water bath with clean water and an antimicrobial agent, agitating gently for 40-60 seconds until a small ice clump remains.
Perform all subsequent steps under sterile conditions in a flow hood, disinfecting the cryovial with 70% ethanol before opening.
Carefully open the disinfected vial and transfer the cell suspension into a 15 ml centrifuge tube containing 8 ml of room-temperature culture medium, mixing gently.
Centrifuge the mixture at 300 x g for 3 minutes to separate the cells and carefully discard the supernatant containing residual freezing medium.
Gently resuspend the cell pellet in 10 ml of fresh culture medium. For adherent cells, divide the suspension between two T25 culture flasks; for suspension cultures, transfer all the medium into one T25 flask to promote effective cell interaction and growth.
Adhere to established subculture protocols for continued growth and maintenance of the cell line, ensuring reliable experimental outcomes.

QUALITY CONTROL AND CRYOPRESERVATION

Sterility

Mycoplasma contamination is excluded using both PCR-based assays and luminescence-based mycoplasma detection methods.
To ensure there is no bacterial, fungal, or yeast contamination, cell cultures are subjected to daily visual inspections.

Freeze medium	As a cryopreservation medium, we use complete growth medium (including FBS) + 10% DMSO for adequate post-thaw viability, or CM-1 (Cytion catalog number 800100), which includes optimized osmoprotectants and metabolic stabilizers to enhance recovery and reduce cryo-induced stress.

Storage conditions

For long-term preservation, place vials in vapor-phase liquid nitrogen at about −150 to −196 °C. Storage at −80 °C is acceptable only as a short interim step before transfer to liquid nitrogen.

Shipping conditions

Cryopreserved cell lines are shipped on dry ice in validated, insulated packaging with sufficient refrigerant to maintain approximately −78 °C throughout transit. On receipt, inspect the container immediately and transfer vials without delay to appropriate storage.

DONOR

Donor age	60 years
Ethnicity	Caucasian

LEGAL AND SAFETY

Safety precautions

When planning to store a cryovial in liquid nitrogen for future thawing, it is mandatory to adhere to stringent safety measures. Appropriate protective gloves and clothing are essential, and the use of a face mask or safety goggles is required during the transfer of frozen samples to or from the liquid nitrogen tank. This is to mitigate the risk of injury from potential cryovial explosions upon removal, which can result in the projection of sharp fragments.

Warranty

At Cytion, we stand behind the quality and viability of our products.
We guarantee that our cells will be viable at the start of culture for
60 days after shipment from our facilities.
We assure that all materials conform to the specifications described in the
Certificate of Analysis and/or Product Sheet.
For full details, please refer to our
Warranty Statement.

Material Transfer Agreement required	Yes