

**4T1 Cells | 300300**

**General information**

**Description**

The 4T1 murine mammary carcinoma cell line is a widely used model in cancer research due to its high similarity to human breast cancer. Derived from a BALB/c mouse, the tumor growth and metastatic spread of the 4T1 cell line closely mimic the behavior of late stage breast cancer in humans. The 4T1 cell line serves as an invaluable tool for studying the progression and metastasis of mammary cancer, including bone metastases and breast cancer metastasis. When injected into BALB/c mice, 4T1 cells spontaneously produce highly metastatic tumors that can spread to various organs such as the lung, liver, lymph nodes, and bone, while the primary tumor continues to grow in situ. This 4T1 syngeneic model is particularly useful for studies of bone metastases and the metastatic phenotype.

The 4T1 cell's utility extends to techniques like bioluminescence imaging, histological analyses, and the use of molecular markers to track the spread and impact of metastatic disease. This approach allows for the examination of spontaneous metastasis from primary tumors to distant organs, aided by techniques like flow cytometry to analyze tumor cells and their receptor expressions. The imagable 4T1 model has enabled biophotonic imaging to track tumor growth and metastasis in vivo in animal models, facilitating studies on metastatic cells in target organs and tumor foci.

The immunocompetent nature of the mouse 4T1 breast tumor cell line allows for investigations into the role of the immune system and immunity in metastasis, as well as immunotherapy of cancer. Moreover, the 4T1 syngeneic tumor model has been instrumental in omics characterization and fusion gene detection.

Overall, the 4T1 mammary carcinoma cell line serves as a versatile tool for studying mammary tumor biology, tumor metastasis, and the development of new treatments in both murine and human contexts.

**Organism** Mouse

**Tissue** Breast, mammary gland

**Disease** Malignant neoplasm

**Applications** 4T1 cells accurately imitate the characteristics of human breast cancer in its most advanced stage - Stage IV.

**Synonyms** 4T1-A, 4T1.0, 4T1/WT

**Characteristics**

**Breed/Subspecies** BALB/cfC3H

**Gender** Female

**Morphology** Epithelial

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**Growth properties** Adherent

**Regulatory Data**

**Citation** 4T1 (Cytion catalog number 300300)

**Biosafety level** 1

**NCBI\_TaxID** 10090

**CellosaurusAccession** CVCL\_0125

**Biomolecular Data**

**Tumorigenic** Yes, in BALB/c mice.

**Handling**

**Culture Medium** RPMI 1640, w: 2.0 mM stable Glutamine, w: 2.0 g/L NaHCO<sub>3</sub> (Cytion article number 820700a)

**Supplements** Supplement the medium with 10% FBS

**Dissociation Reagent** Accutase

**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.

**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.

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### Thawing and Culturing Cells

1. Confirm that the vial remains deeply frozen upon delivery, as cells are shipped on dry ice to maintain optimal temperatures during transit.
2. Upon receipt, either store the cryovial immediately at temperatures below  $-150^{\circ}\text{C}$  to ensure the preservation of cellular integrity, or proceed to step 3 if immediate culturing is required.
3. For immediate culturing, swiftly thaw the vial by immersing it in a  $37^{\circ}\text{C}$  water bath with clean water and an antimicrobial agent, agitating gently for 40-60 seconds until a small ice clump remains.
4. Perform all subsequent steps under sterile conditions in a flow hood, disinfecting the cryovial with 70% ethanol before opening.
5. 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.
6. Centrifuge the mixture at  $300 \times g$  for 3 minutes to separate the cells and carefully discard the supernatant containing residual freezing medium.
7. 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.
8. Adhere to established subculture protocols for continued growth and maintenance of the cell line, ensuring reliable experimental outcomes.

### Incubation Atmosphere

$37^{\circ}\text{C}$ , 5%  $\text{CO}_2$ , humidified atmosphere.

### Flask Coating

None

### Shipping Conditions

Cryopreserved cell lines are shipped on dry ice in validated, insulated packaging with sufficient refrigerant to maintain approximately  $-78^{\circ}\text{C}$  throughout transit. On receipt, inspect the container immediately and transfer vials without delay to appropriate storage.

### Storage Conditions

For long-term preservation, place vials in vapor-phase liquid nitrogen at about  $-150$  to  $-196^{\circ}\text{C}$ . Storage at  $-80^{\circ}\text{C}$  is acceptable only as a short interim step before transfer to liquid nitrogen.

## Quality Control & Molecular Analysis

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### **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.