

## BT-549/RFP Cell Line

**CATALOG NUMBER:** AKR-255

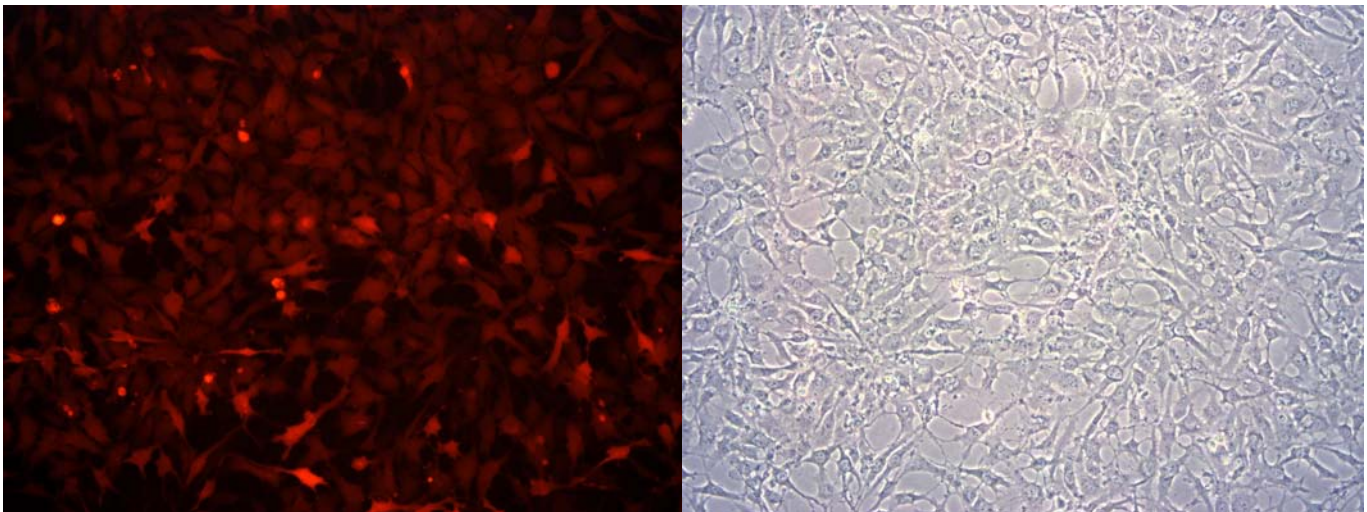
**STORAGE:** Liquid nitrogen

*Note: For best results begin culture of cells immediately upon receipt. If this is not possible, store at -80°C until first culture. Store subsequent cultured cells long term in liquid nitrogen.*

**QUANTITY & CONCENTRATION:** 1 mL,  $1 \times 10^6$  cells/mL in 70% DMEM, 20% FBS, 10% DMSO

### **Background**

The BT-549 cell line was isolated in 1978 by W.G. Coutinho and E.Y. Lasfargues. Source tissue consisted of a papillary, invasive ductal tumor which had metastasized to 3 of 7 regional lymph nodes. The established population was polymorphic with epithelial like components and multinucleated giant cells. A mucin-like material was secreted into the medium. *In vitro*, the BT-549 cell line has abundant activity in both the Boyden chamber chemoinvasion and chemotaxis assay. The BT-549 cell line is also able to grow on agarose, an indicator of transformation and tumorigenicity, and displays a relatively high colony forming efficiency. Our BT-549/RFP cell line stably expresses RFP and Puromycin resistant genes that were introduced using lentivirus.



**Figure 1.** BT-549/RFP Cell Line. Left: RFP Fluorescence; Right: Phase Contrast.

### **Quality Control**

This cryovial contains at least  $1.0 \times 10^6$  BT-549/RFP cells as determined by morphology, trypan-blue dye exclusion, and viable cell count. The BT-549/RFP cells are tested free of microbial contamination.

## **Medium**

1. Culture Medium: D-MEM (high glucose), 10% fetal bovine serum (FBS), 0.1 mM MEM Non-Essential Amino Acids (NEAA), 2 mM L-glutamine, 1% Pen-Strep.
2. Freeze Medium: 70% DMEM, 20% FBS, 10% DMSO.

## **Methods**

### **Establishing BT-549/RFP Cultures from Frozen Cells**

1. Place 10 mL of complete DMEM growth medium in a 50-mL conical tube. Thaw the frozen cryovial of cells within 1–2 minutes by gentle agitation in a 37°C water bath. Decontaminate the cryovial by wiping the surface of the vial with 70% (v/v) ethanol.
2. Transfer the thawed cell suspension to the conical tube containing 10 ml of growth medium.
3. Collect the cells by centrifugation at 1000 rpm for 5 minutes at room temperature. Remove the growth medium by aspiration.
4. Resuspend the cells in the conical tube in 15 mL of fresh growth medium by gently pipetting up and down.
5. Transfer the 15 mL of cell suspension to a T-75 tissue culture flask. Place the cells in a 37°C incubator at 5% CO<sub>2</sub>.
6. Monitor cell density daily. Cells should be passaged when the culture reaches 95% confluence.

## **Warranty**

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