

Research Use Only. Not for use in diagnostic procedures.

Bioware[®] Brite Cell Line 4T1-Red-FLuc-GFP

Product No.: BW128090

**We recommend using this line in Nude and SCID mouse models.
An immune response may occur in Balb/c mice.**

Material Provided

Cells: 2 x 1 mL frozen aliquots (BW128090V)

Format: 1.0 x 10⁶ cells / mL in 95% FBS, 5% DMSO

DESIGNATION	4T1-Red-FLuc-GFP
Tissue	Mouse: mammary gland; adenocarcinoma
Source of Parental Line	ATCC (CRL-2539)
Gene Transfer Vehicle	Red-FLuc-GFP 3d generation lentivirus
Bioluminescence In Vitro	At least 15,000 photons/cell/sec. Exact number will vary depending on imaging and culturing conditions.
Recommended Media and FBS	RPMI 1640 ATCC Cat. No. 30-2001. Supplement the above with 10% Hyclone Fetal Bovine Serum (FBS) GE HealthCare Cat. No. SH300071
Recommended Storage Conditions	Remove frozen cells from dry ice packaging and immediately place cells at a temperature below -130° C, preferably in liquid nitrogen vapor, until ready to use.
Average Doubling Time	14 hours
Other Recommendations	When initially thawing, use T25 flask or 10cm plate. Cells should be ready to expand within 1-4 days. Please note there is NO antibiotic selection for this cell line. Refer to Cell Culture Guidelines for more detailed instructions.

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The Features

Perkin Elmer Bioware® Ultra cell line models offer researchers the ability to:

- Monitor early tumor development
- Monitor tumor growth and metastases *in vivo*
- Quantify tumor burden in the whole animal
- Follow responses to therapeutic treatments non-invasively in longitudinal studies using the same cohorts of mice
- Perform *in vitro* and *ex vivo* analysis with GFP

Murine Pathogen Free

All Perkin Elmer cell lines are confirmed to be pathogen free by the IMPACT Profile I (PCR) at the University of Missouri Research Animal Diagnostic and Investigative Laboratory.

Cell Line Stability

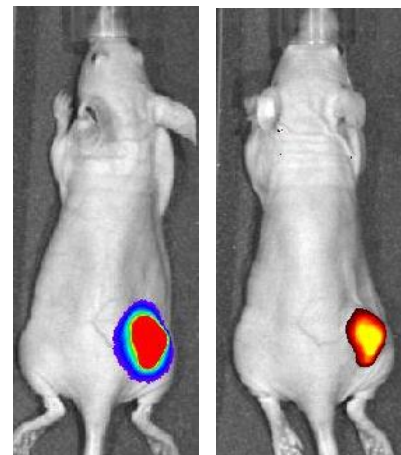
Cell may undergo genotypic changes resulting in reduced responsiveness over time in normal cell culture conditions. Genetic instability is a biological phenomenon that occurs in all stably transfected cells. Therefore, it is recommended to prepare an adequate number of frozen stock at early passages.

Product Warranty

PerkinElmer warrants that cells will be viable upon shipment from PerkinElmer for a period of thirty days, provided they have been properly stored and handled during this period.

Mouse Mammary Gland Cancer Cell Line: 4T1-Red-FLuc-GFP

4T1-Red-FLuc-GFP is a luciferase expressing cell line which was been stably transfected with the firefly luciferase gene from *Luciola Italica* (Red-FLuc) fused to GFP protein. The cell line was established by transducing lentivirus containing Red-FLuc luciferase-T2A-GFP gene under the control of human ubiquitin C promoter. These cells will serve as a new tool to detect drug efficacy *in vitro* and *in vivo* with high sensitivity in Nude and SCID mouse models.



Bioluminescence and GFP images of 4T1 –Red-FLuc-GFP subcutaneous tumors in nude mice

Growth Curve of 4T1-Red-FLuc-GFP Cells

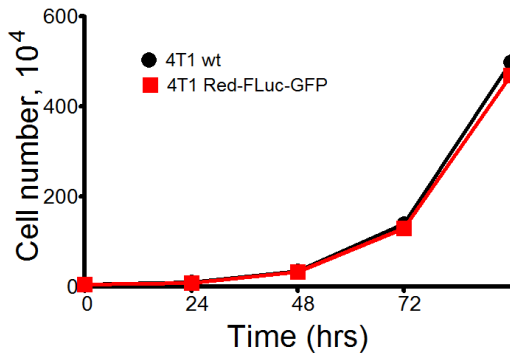


Figure 1. 5×10^4 cells were plated on 6cm plate and the total numbers of cells were counted every 24 h using a Nexcelom automatic cell counter.

In Vitro BLI Signal Stability

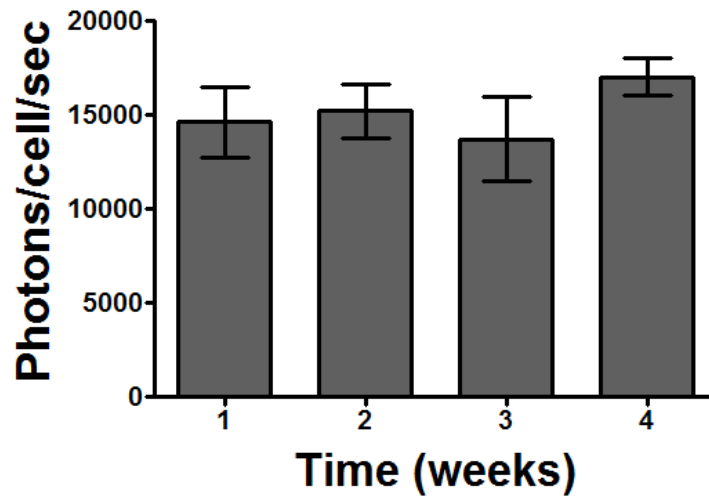
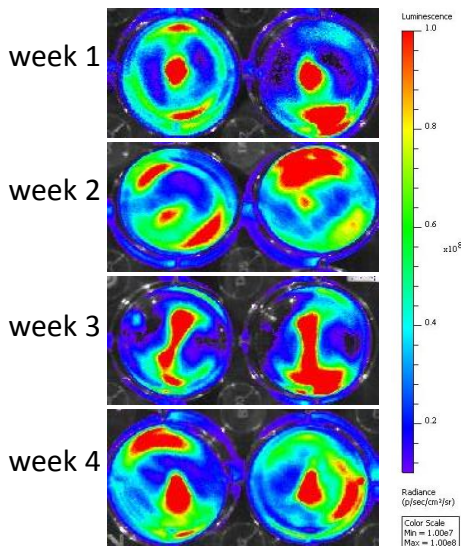


Figure 2. 5×10^4 cells were plated per well in 24-well plates. Cells were incubated at 37 °C for recovery overnight and luciferase assay was performed using the PerkinElmer IVIS® SpectrumCT. Each experiment was done in quadruplicates. The cells were maintained in continuous culture over four weeks and weekly luciferase assay was performed. Bioluminescence data was analyzed using the Living Image 4.0 software.

Subcutaneous Tumor Growth in a Nu/nu Mouse

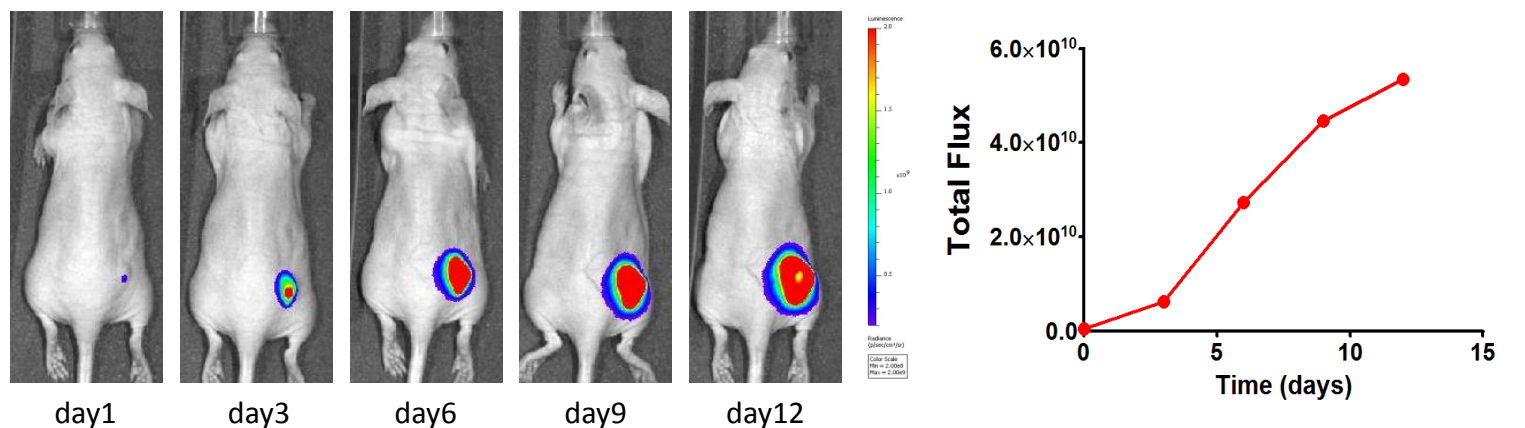


Figure 3. 1×10^6 4T1-Red-FLuc-GFP cells were injected subcutaneously into the dorsal region near the thigh of female nu/nu mouse. Tumor growth was monitored for luciferase expression using the PerkinElmer IVIS[®] Spectrum at various time points. For luciferase expression, mice were imaged 10 minutes post i.p. injection of luciferin at 150mg/kg at various time points. The image above shows tumor growth from representative mouse.

For more information on our *in vivo* imaging agents, please visit our website: www.perkinelmer.com/bioware.

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