

Cell name: A-431 x ABCG2-Neo  
Cat. No. A-0310

### Product description

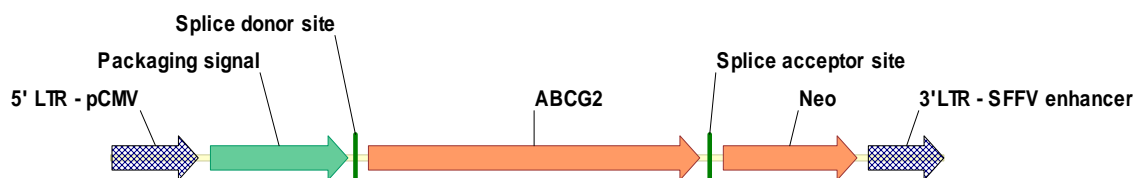
A-431 x ABCG2-Neo was established by retroviral transduction of human ABCG2 (ATP-binding cassette, sub-family G, member 2) cDNA, along with the Neomycin resistance gene, into A-431 cells and flow cytometry sorting of the anti-Human ABCG2 immunolabeled population.

### Transgene construct information

#### Expression cassette type

Single-promoter, monocistronic

#### Expression cassette map



#### Expression cassette features

| Element                  | Type                          | Species      | RefSeq  | Mutation / Discrepancy |
|--------------------------|-------------------------------|--------------|---|------------------------|
| SFFV                     | promoter, viral, constitutive | -            | -   | -                      |
| Splice donor site        | retroviral                    | -            | -   | -                      |
| ABCG2                    | CDS                           | Homo sapiens | <a href="http://www.ncbi.nlm.nih.gov/RefSeq/record/NM_004827.2">NM_004827.2</a> | none / none            |
| Splice acceptor site     | retroviral                    | -            | -   | -                      |
| Neomycin resistance gene | CDS                           | -            | -   | none / none            |

### Transgene protein information

The transmembrane protein encoded by this gene is included in the superfamily of ATP-binding cassette (ABC) transporters. ABC proteins transport various molecules across extra- and intra-cellular membranes. ABC genes are divided into seven distinct subfamilies (ABC1, MDR/TAP, MRP, ALD, OABP, GCN20, White). This protein is a member of the White subfamily. Alternatively referred to as a breast cancer resistance protein, this protein functions as a xenobiotic transporter which may play a major role in multi-drug resistance. It likely serves as a cellular defense mechanism in response to mitoxantrone and anthracycline exposure. Significant expression of this protein has been observed in the placenta, which may suggest a potential role for this molecule in placenta tissue. Multiple transcript variants encoding different isoforms have been found for this gene. [Source: NCBI Gene, <http://www.ncbi.nlm.nih.gov/gene/9429>]

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## Cell culture characteristics

### Source

Human skin epidermoid carcinoma

### Morphology

Epithelial

### Growth properties

Adherent

### Culture conditions

Culture cells at 37°C in humidified atmosphere with 5% CO<sub>2</sub>. The base medium for this cell line is α-MEM with 4.5 g/l glucose. To make the complete growth medium, add fetal bovine serum to a final concentration of 10%.

### Subculturing

Remove culture medium, rinse twice thoroughly with PBS, and detach cells by treating with 0.04 mL/cm<sup>2</sup> of 0.25% trypsin / 1 mM EDTA solution for 15 minutes at 37°C. Add 5x volume of complete medium to neutralize trypsin-EDTA, pellet cells at 250 x g, discard supernatant, and resuspend in culture medium. Subcultivate in a ratio of 1:4-1:6 three times a week. Recommended plating density: 3x10<sup>4</sup> cells/cm<sup>2</sup>.

### Preservation

Freeze Medium: Complete growth medium with 10% DMSO.

Storage Temperature: Liquid nitrogen vapour phase.

### Population doubling time

~30 hours

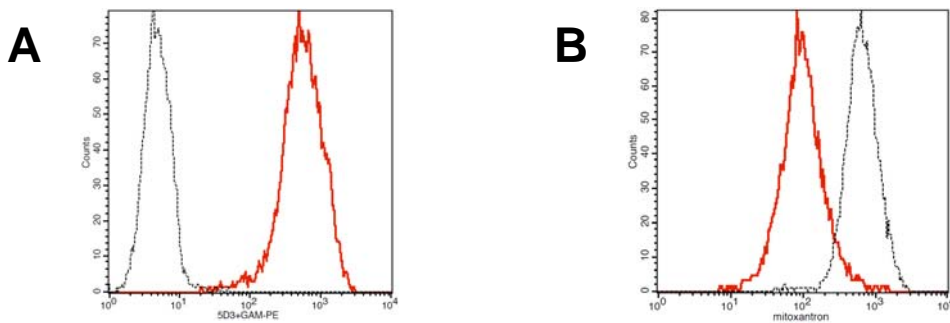
### Sterility testing

Mycoplasma: negative

### Comment

This cell line overexpresses Epidermal Growth Factor Receptor (EGFR).

## Validation results



**A:** Red line: A-431 x ABCG2-Neo cells incubated with anti-Human ABCG2 antibody.

Dotted line: A-431 x ABCG2-Neo cells incubated with isotype control.

Indirect immunofluorescent labeling with anti-Human ABCG2 (clone 5D3) antibody (R&D Systems®) or isotype control + PE labeled anti-Mouse IgG2b secondary antibody. Detection by flow cytometry.

**B:** Red line: A-431 x ABCG2-Neo cells incubated with mitoxantrone.

Dotted line: A-431 x ABCG2-Neo cells incubated with mitoxantrone and Ko143.

Mitoxantrone uptake detected by flow cytometry at 633 nm in the presence or absence of Ko143, a specific inhibitor of ABCG2.

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