

Cell name: A-431 x ABCB1-Neo M3 (low)
Cat. No. A-0361

Product description

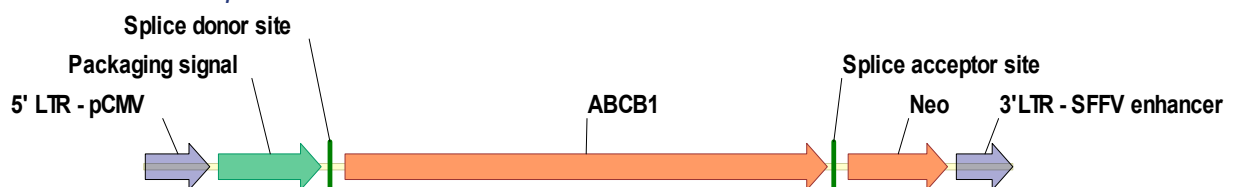
A-431 x ABCB1-Neo, clone M3, was established by retroviral transduction of human ABCB1 (also known as P-glycoprotein or MDR1) cDNA into A-431 cells and limiting dilution cloning. The clone M3 exhibits relatively low ABCB1 transport activity based on calcein uptake assay.

Transgene construct information

Expression cassette type

Single-promoter, bicistronic

Expression cassette map



Expression cassette features

Element	Type	Species	RefSeq	Mutation / Discrepancy
SFFV	promoter, viral, constitutive	-	-	-
Splice donor site	Splice donor site	-	-	-
ABCB1	CDS	Homo sapiens	AF016535	none / none
Splice acceptor site	Splice acceptor site	-	-	-
Neomycin resistance gene	CDS	-	-	none / none

Transgene protein information

The transmembrane protein encoded by this gene is a member of 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 MDR/TAP subfamily. Members of the MDR/TAP subfamily are involved in multidrug resistance. The protein encoded by this gene is an ATP-dependent drug efflux pump for xenobiotic compounds with broad substrate specificity. It is responsible for decreased drug accumulation in multidrug-resistant cells and often mediates the development of resistance to anticancer drugs. This protein also functions as a transporter in the blood-brain barrier. [Source: NCBI Gene, <http://www.ncbi.nlm.nih.gov/gene/5243>]

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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% CO₂. 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/cm² 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: 3x10⁴ cells/cm².

Preservation

Freeze Medium: Complete growth medium with 10% DMSO.

Storage Temperature: Liquid nitrogen vapour phase.

Population doubling time

~30 hours

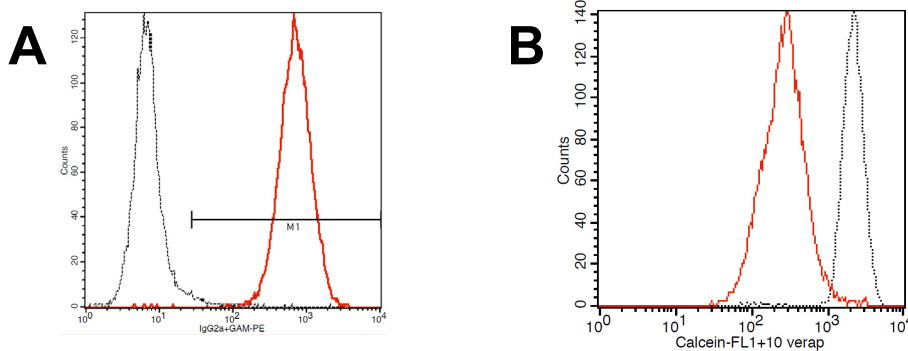
Sterility testing

Mycoplasma: negative

Comment

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

Validation results



A: Red line: A-431 x ABCB1-Neo M3 cells incubated with anti-Human ABCB1 antibody.

Dotted line: A-431 x ABCB1-Neo M3 cells incubated with isotype control.

Indirect immunofluorescent labeling with anti-Human ABCB1 antibody (clone MRK16, Alexis Biochemicals / Enzo Life Sciences) or isotype control + PE labeled anti-Mouse IgG secondary antibody.

Detection by flow cytometry.

B: Red line: A-431 x ABCB1-Neo M3 cells incubated with calcein.

Dotted line: A-431 x ABCB1-Neo M3 cells incubated with calcein and verapamil.

Calcein uptake was detected by flow cytometry at 485 nm in the presence or absence of verapamil, an inhibitor of ABCB1.

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