

A375-GFP | 305665

General information

Description

A375-GFP is a genetically engineered variant of the human malignant melanoma cell line A375, stably expressing enhanced green fluorescent protein (eGFP). The parental A375 cell line is derived from a skin melanoma tumor in an adult patient and is widely used as a model for cutaneous melanoma, particularly for studies involving oncogenic BRAF signaling, as it harbors the BRAF V600E mutation. This mutation leads to constitutive activation of the MAPK/ERK pathway, driving proliferation and survival, and making A375 cells highly relevant for investigating targeted therapies such as BRAF and MEK inhibitors. The GFP-expressing derivative retains these molecular and phenotypic characteristics while enabling fluorescence-based applications.

The stable incorporation of the eGFP reporter allows for real-time visualization of A375-GFP cells in both in vitro and in vivo systems. Fluorescence imaging facilitates monitoring of cell proliferation, migration, invasion, and morphological changes, as well as tracking tumor growth and metastatic dissemination in xenograft models. The enhanced GFP variant provides improved brightness and stability compared to earlier GFP constructs, enabling sensitive detection even at low cell numbers. This makes A375-GFP particularly useful in co-culture experiments, high-content imaging platforms, and studies requiring precise spatial resolution of tumor cell behavior.

A375-GFP maintains the aggressive and proliferative phenotype of the parental melanoma line, including responsiveness to MAPK pathway inhibitors and the capacity for invasion and metastasis in experimental models. The addition of GFP expands its utility for drug screening, live-cell imaging, and tumor-microenvironment interaction studies. As with other reporter-labeled cell lines, validation of fluorescence stability and consistency across passages is recommended for specific experimental applications.

Genetic modification: Stably modified by replication-incompetent lentiviral transduction to express the ZsGreen1 green fluorescent protein reporter; maintained as a polyclonal population under puromycin selection (1–5 µg/mL). S1/BSL-1 containment.

Organism Human

Tissue Leg, skin

Disease Amelanotic melanoma

Characteristics

Age 54 years

Gender Female

Ethnicity Caucasian

Growth properties Adherent

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Regulatory Data

Citation	A375-GFP (Cytion catalog number 305665)
Biosafety level	1
NCBI_TaxID	9606
CellosaurusAccession	CVCL_QZ67
GMO Status	GMO-S1: This cell line contains a stably integrated ZsGreen1 green fluorescent protein reporter introduced via replication-incompetent lentiviral transduction. The resulting polyclonal cell population was maintained under puromycin selection (1–5 µg/mL). S1 containment is required. This classification applies only within Germany and may differ elsewhere.

Biomolecular Data

Antigen expression	ZsGreen1 (green fluorescent protein)
Mutational profile	Mutation: BRAF, Simple, p.Val600Glu (c.1799T>A), Homozygous (from parent cell line).Mutation, CDKN2A, Simple, p.Glu61Ter (c.181G>T) (p.Gly75Val, c.224G>T), Homozygous (from parent cell line).Mutation, CDKN2A, Simple, p.Glu69Ter (c.205G>T) (p.Gly83Val, c.248G>T), Homozygous (from parent cell line).Mutation, TERT, Simple, c.1-146C>T (c.250C>T) (C250T), Unspecified, Note=In promoter (from parent cell line).

Handling

Culture Medium	DMEM, w: 4.5 g/L Glucose, w: 4 mM L-Glutamine, w: 3.7 g/L NaHCO ₃ , w: 1.0 mM Sodium pyruvate (Cytion article number 820300a)
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.

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Seeding density 1 to 3×10^4 cells/cm²

Fluid renewal 2 to 3 times per week

Freeze medium As a cryopreservation medium, we use complete growth medium + 10% DMSO for adequate post-thaw viability.

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°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°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 200 x g for 5 minutes, carefully discard the supernatant containing freezing medium.
7. Follow the procedure described under Post-Thaw Recovery

Incubation Atmosphere 37°C, 5% CO₂, humidified atmosphere.

Shipping Conditions

Cryopreserved cell lines are shipped on dry ice in validated, insulated packaging with sufficient refrigerant to maintain approximately -78 °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 °C. Storage at -80 °C is acceptable only as a short interim step before transfer to liquid nitrogen.

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Quality Control & Molecular Analysis