

Green-AIDeSense™ ALDH1A1 Live Cell Dye

Live Cell Dye

Cat. # SCT065

pack size: 1 Kit

FOR RESEARCH USE ONLY.
NOT FOR USE IN DIAGNOSTIC PROCEDURES.
NOT FOR HUMAN OR ANIMAL CONSUMPTION.

Store at -20°C



Data Sheet

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Background

Cancer stem cells (CSCs) have an increased capacity to activate antiapoptotic/prosurvival pathways as well as overexpressing ATP-binding cassette transporters which act as potent efflux pumps for potential therapeutics.

Aldehyde dehydrogenases (ALDH), in particular the ALDH1A1 isoform, is believed to be a reliable marker of CSCs across many cancer types, including prostate, lung, breast, esophageal, and ovarian cancers. ALDH1A1 is associated with increased chemotherapy resistance and poor clinical outcome and is as an established stem cell marker.

Current methods to identify ALDH is living cells are limited by cross-reactivity with several ALDH isoforms producing difficulties in data analysis and prognosis. Chan, et al. have created an ALDH1A1 isoform specific live cell dye termed Green-AIDeSense™ which labels cancer stem cells and exhibits a 20-fold fluorescent enhancement when bound to ALDH1A1. The dye identifies ALDH1A1 positive CSCs using live cell imaging, flow cytometry and *in vivo* studies.

Spectral Properties: abs_{max} : 496 nm, em_{max} : 516 nm.

Kit Components

The Green-AIDeSense™ ALDH1A1 Live Cell Dye Kit (SCT065) contains:

- 1) Ctrl-Green-AIDeSense™ Dye (CS226256), 100 µg. Store at -20°C. Protect from light. MW: 380.3 g/mol.
- 2) Green-AIDeSense™ Dye (CS226257), 100 µg. Store at -20°C. Protect from light. MW: 438.4 g/mol.

Quality Control

Color: Orange

Form: Solid

Solubility: DMSO: >10 mg/mL

Purity: ≥ 80.0%

Elemental Analysis/Identification: Conforms

Instructions For Use

Reconstitute the Green-AIDeSense™ and Ctrl-Green-AIDeSense™ dye in DMSO at 2 mM and freeze stock aliquots at -20°C. Avoid multiple freeze thaw cycles.

Live Cell Imaging Protocol

1. Seed cells onto a tissue culture treated plate and grow to 70-80% confluence at 37°C, 5% CO₂ in standard growth media (i.e. DMEM + 10% FBS).
2. Remove media and treat cells with 2 µM Green-AIDeSense™ or Ctrl-Green-AIDeSense™ dye diluted in standard growth media.
3. Incubated cells 30 minutes at room temperature.
4. Immediately image stained cells on an epifluorescence microscope using a GFP filter set.

Flow Cytometry Protocol

1. Harvest cells from culture and resuspended in ice-cold FACS buffer (1x PBS + 2% BSA, 2 mM EDTA).
2. Incubate 100,000 cells for 30 minutes on ice with 2 µM Green-AIDeSense™ or Ctrl-Green-AIDeSense™ dye.
3. Pellet samples and then resuspended cells in ice-cold FACS buffer. Place resuspended cells (200 µL/well) in a 96-well microplate for flow analysis.
4. Samples were gated according to recommended parameters and analyzed with green fluorescence channel on Guava easyCyte HT8 flow cytometer.

References

- 1) Chan, et al. Surveillance of Cancer Stem Cell Plasticity Using an Isoform-Selective Fluorescent Probe for Aldehyde Dehydrogenase 1A1. ACS Cent Sci. 2018 Aug 22;4(8):1045-1055.

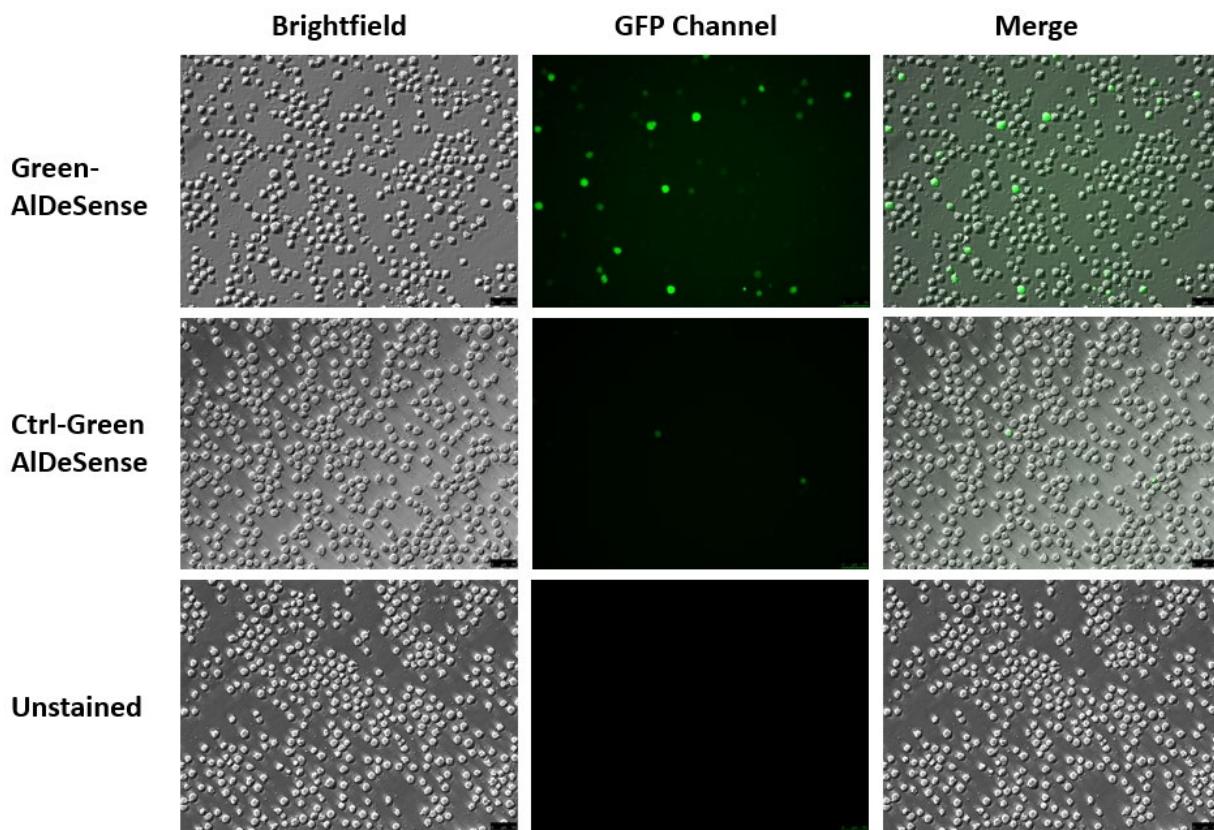


Figure 1. Live cell imaging of ALDH1A1 using Green-AIDeSense™. Green-AIDeSense™ identifies ALDH1A1(+) population in the human blood chronic myelogenous leukemia K562 cell line. Ctrl-Green-AIDeSense™ shows little to no staining and has a similar staining profile as the unstained control.

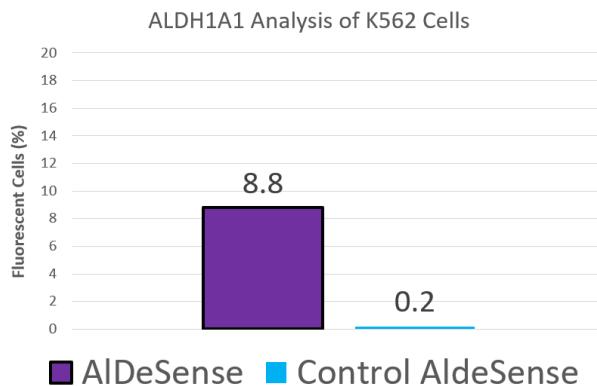


Figure 2. Quantification of ALDH1A1(+) K562 cells using Green-AIDeSense™. Green-AIDeSense™ stains roughly 9% of ALDH1A1(+) K562 cells in a live cell imaging experiment while the Ctrl-Green-AIDeSense™ stains less than 0.2%.

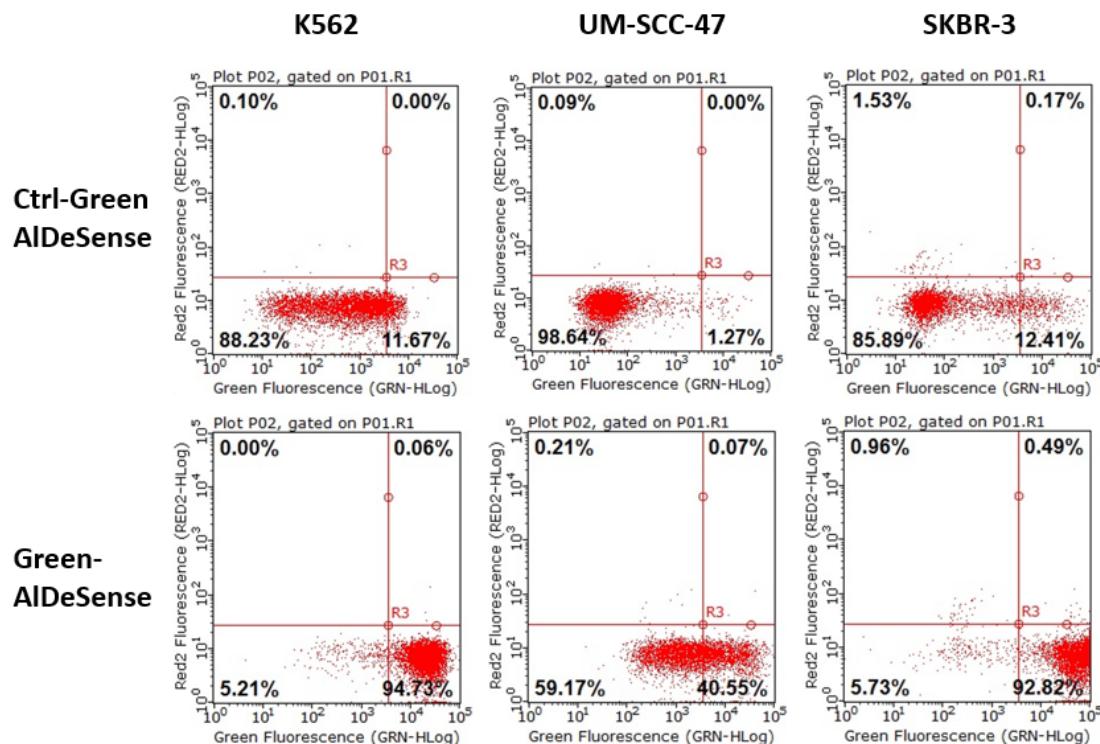


Figure 3. Flow analysis of ALDH1A1 in cancer cells using Green-AIDeSense™. K562 (human leukemia), UM-SCC-47 (human squamous cell carcinoma) and SKBR-3 (human breast cancer) cell lines were analyzed for ALDH1A1 expression using flow cytometry.

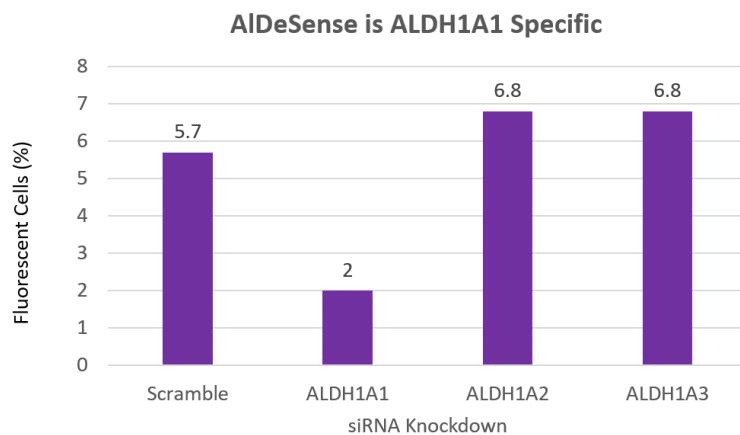


Figure 4. Green-AIDeSense™ shows ALDH1A1 isoform specificity. K562 cells were treated with siRNAs for ALDH1A1, ALDH1A2 and ALDH1A3 isoforms and then stained with Green-AIDeSense™. Signal only decreased in the ALDH1A1 knockdown after siRNA incubation suggesting ALDH1A1 isoform specificity of the dye.