Data Sheet

BioTracker™ Far-Red Peroxynitrite Live Cell Dye

Live Cell Probe

SCT052

Pack Size: 1 mg

Store at −20 °C

FOR RESEARCH USE ONLY

Not for use in diagnostic procedures. Not for human or animal consumption.

Background

Reactive oxygen species (ROS) and reactive nitrogen species (RNS) have been linked to a variety of physiological and pathological outcomes. Peroxynitrite (ONOO-) has garnered a lot of attention due to its key role in signal transduction and antimicrobial activities. It is endogenously produced by the reaction of nitric oxide (NO) and superoxide radical anion (O_2^-), however, peroxynitrite is more cytotoxic than NO or O_2^- which appears to contribute to significant pathologies such as cardiovascular disease, diabetes, and cancer. Peroxynitrite's powerful oxidizing capabilities also contribute to various intracellular reactions that result in cell apoptosis or necrotic cell death. Understanding the various cellular roles and capabilities of ONOO- may help to reduce related pathologies. Peroxynitrite is unstable and highly reactive, giving it a short cellular lifespan. This makes it nearly impossible to detect using traditional analytical methods.

The BioTracker™ Far-Red Peroxynitrite Live Cell Dye can be used for ONOO- detection. The Far-Red Peroxynitrite probe is cell permeable and demonstrates high sensitivity and selectivity for peroxynitrite over other ROS/RNS. The probe is activated via the reaction of a hydrazide group in the presence of ONOO-. This probe can be used to detect exogenous and endogenous peroxynitrite in living systems and displays long wavelength absorption and emission bands.

Source

The BioTracker™ Far-Red Peroxynitrite Live Cell Dye (SCT052) does not contain genetically modified organisms.

Spectral Properties

Excitation max: 600 nm

Emission max: 638 nm

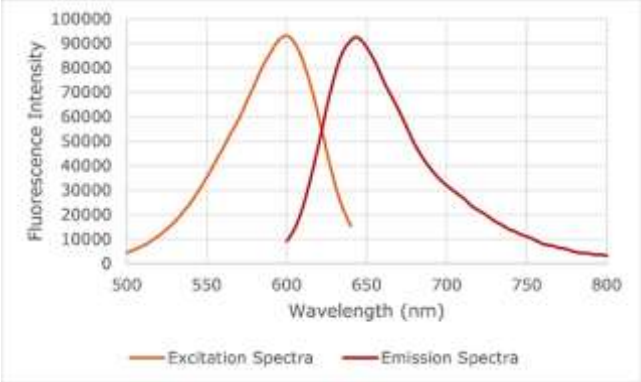

Figure 1: Probe excitation and emission data. 7 µL of probe at stock concentration (10 mM) was diluted in 1 mL of Tris HCl buffer (10 µM, pH 7.0). Peroxynitrite (516620) was added (500 µM) to induce probe activation before undergoing excitation and emission scans. Spectral scans were conducted using a PerkinElmer FL8500 Fluorescence Spectrophotometer.

Quality Control Testing

Purity: ≥ 98% confirmed by HNMR, LC-MS and HPLC and elemental analysis.

Molar Mass: 480.60 g/mol

Storage and Handling

Store BioTracker™ Far-Red Peroxynitrite Live Cell Dye at −20 °C, desiccate and protect from light.

Note: Centrifuge vial briefly to collect contents at bottom of vial before opening.

Presentation

Lyophilized

Representative Data

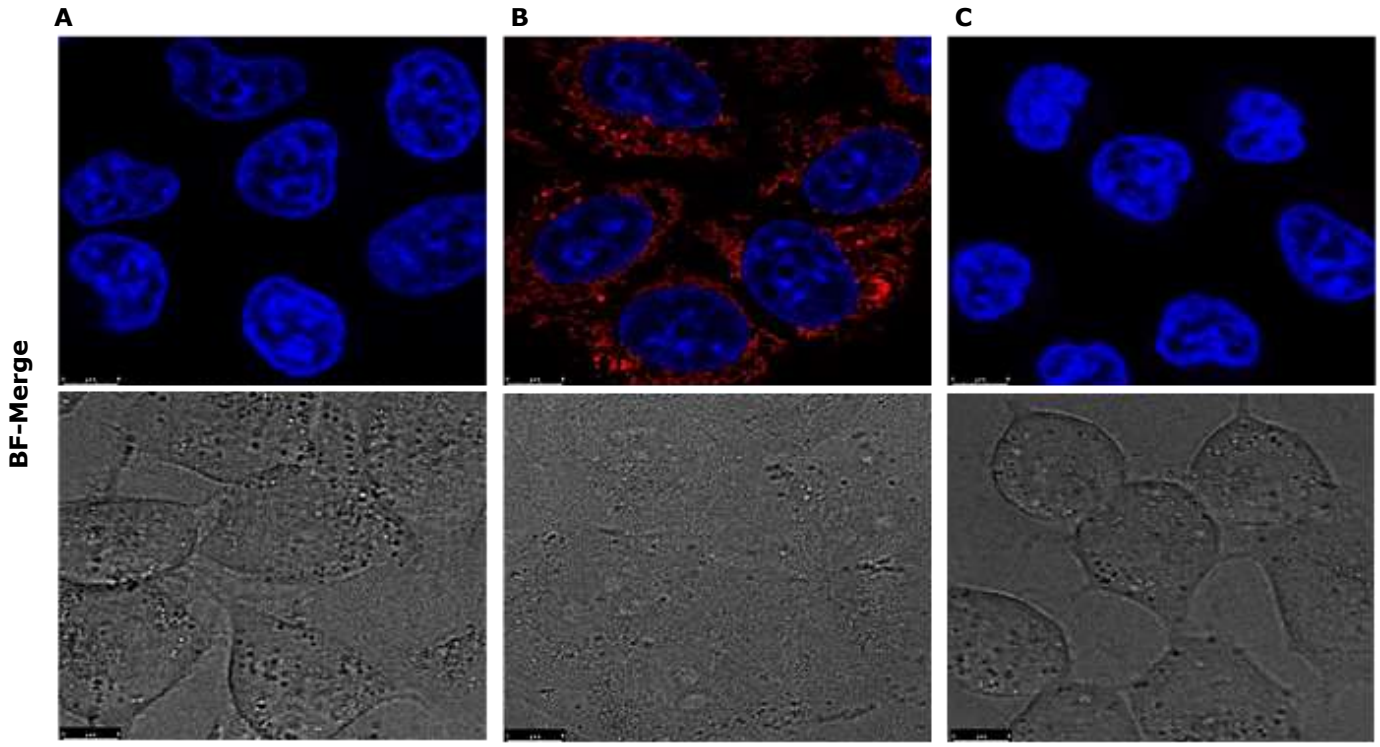

Figure 3: (**A**) Imaging of exogenous peroxynitrite (ONOO**-**) in HeLa cells. Cells were treated with 10 µM peroxynitrite dye for 30 minutes to reveal basal ONOO**-** levels. (**B**) Cells were pre-treated with a peroxynitrite generator (SIN-1) at 50 µM before being stained with 10 µM far-red peroxynitrite live cell dye for 30 minutes. (**C**) Cells were pre-treated with a peroxynitrite scavenger, ebselen, before being incubated with 10 µM peroxynitrite probe.

Protocols

Reagent Preparation

1. Before opening the vial, spin down the solid to the bottom by a microcentrifuge or by a desktop centrifuge.
2. Warm the vial to the room temperature and add DMSO to make a 1000X stock solution of 5-10 mM (freeze aliquots at −20 °C).
3. Dilute in cell culture media at a final concentration of 5-10 µM and add to cells in culture. Incubate at 37 °C for 30 minutes.
4. Wash cells with PBS buffer before imaging.

   Note: Optimal concentration must be determined by end user.

References

1. Wu D, Ryu J-C, Chung YW, Lee D, Ryu J-H, Yoon J-H, Yoon J. 2017. A Far-Red-Emitting Fluorescence Probe for Sensitive and Selective Detection of Peroxynitrite in Live Cells and Tissues. Analytical Chemistry. 89(20):10924–10931. doi:https://doi.org/10.1021/acs.analchem.7b02707.

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