LYSO-ID® Red cytotoxicity kit (GFP-CERTIFIED®)

A Rapid, Quantitative and HTS- compatible Live Cell Cytotoxicity Assay

The LYSO-ID® Red Cytotoxicity Kit monitors dysfunction of lysosomal degradation using a drug-like cationic amphiphilic tracer (CAT) dye that rapidly and selectively stains acidic organelles, and is suitable for monitoring accumulation of lysosomes and lysosome-like structures in live cells.

Mechanism of Action
A cationic amphiphilic tracer (CAT) dye that rapidly partitions into cells in a similar manner as drugs that induce phospholipidosis. This tracer was created by placing titratable groups on the probe (dye), to enable labeling to be expanded into lamellar inclusion bodies of cells pretreated with weakly basic cell-permeant compounds, such as the antimalarial drug chloroquine. Besides lysosomes themselves, the probe can be employed for highlighting lysosome-like organelles under conditions wherein cells produce vacuoles that contain most of the degradative enzymes of the lysosome but are not as acidic as the parent organelle.

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Ordering Information

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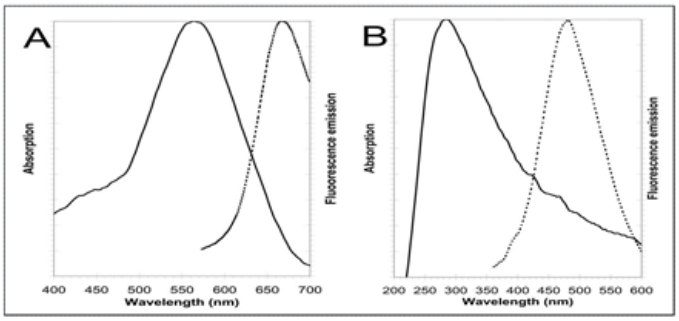
ENZ-51015-KP002	1Kit

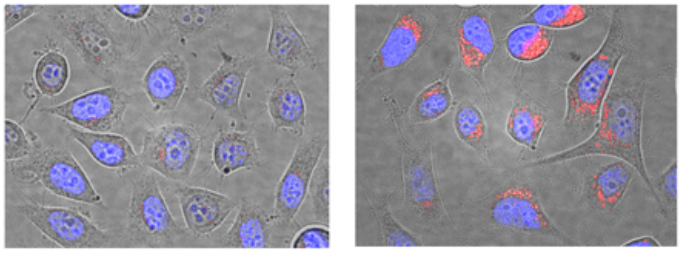
Manuals, SDS & CofA

View Online »

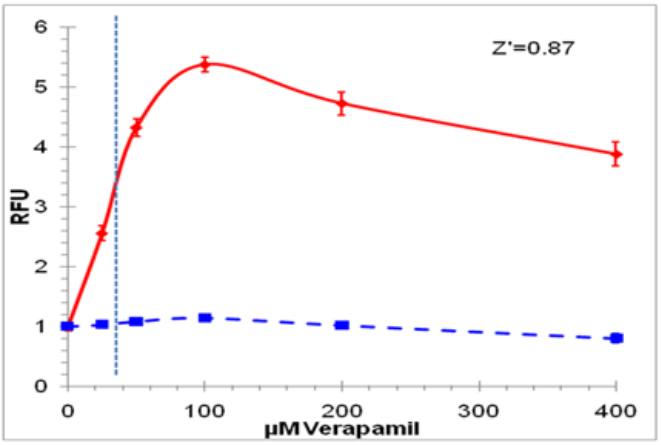
- Assay includes unique drug-like dye that rapidly partitions into cells and labels acidic organelles
- Only commercial assay available that allows for long-term cell monitoring of cytotoxic effects
- Multi-well, high-throughput with rapid 10-15 minute dye incubation
- No co-incubation with artificial phospholipid analogs required for detection, eliminating the potential for confounding dye-associated artifacts
- Monitors lysosome accumulation as a response to prolonged drug treatment
- Quantitative results in as little as 3 hours, including drug treatment



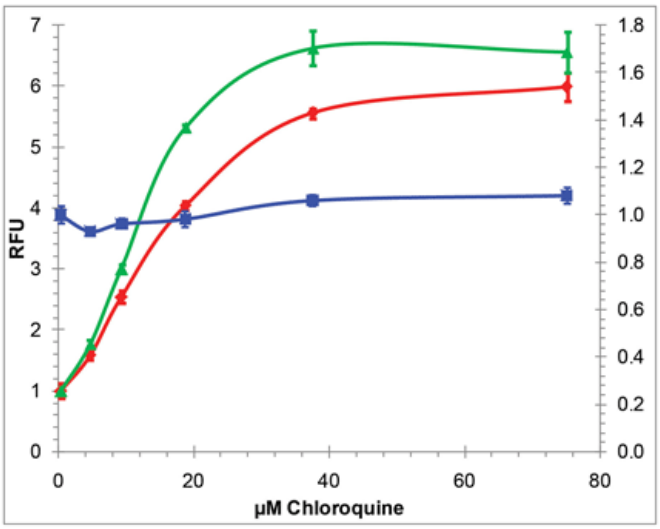
Excitation and emission spectra for A) LYSO-ID® Red Detection Reagent and B) Hoechst 33342 Nuclear Stain.



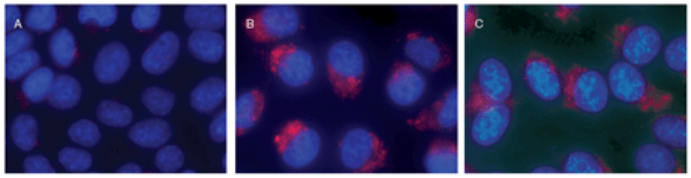
Composite bright-field and fluorescence microscopy images of control U2-OS cells (left) and cells pre-treated with 64µM Chloroquine for 5 hours (right). Cells were stained with LYSO-ID® Red dye for 10 minutes. Nuclei were counter-stained with Hoechst 33342 dye.



High Throughput Screening of Therapeutics for Lysosome-perturbing Activity. Toxicity of Verapamil in U-2 OS cells was estimated using a conventional fluorescence microplate reader. U-2 OS cells were treated with Verapamil for 18 hours and stained with LYSO-ID® Red dye for 15 minutes. The high Z-factor (0.87 for 100 μM Verapamil) indicates that LYSO-ID® Red dye is suitable for HTS applications. Hoechst is used as a counterstain as a normalization control for cell number.



Eliminate Confounding Dye-associated Artifacts. The short 15-minute LYSO-ID® Red dye incubation eliminates the potential for confounding dye-associated artifacts. Relative fluorescent intensity of U-2 OS cells treated with chloroquine at different concentrations for 24 h. Cells stained with LipidTox dye (green line) were incubated in the presence of the fluorescent lipid for 24 h during treatment with the drugs. Cells stained with LYSO-ID® Red dye (red line) or Hoechst 33342 (blue line) were stained for 15 min after drug incubation.



Drug-induced lysosome accumulation in U-2 OS cells was evaluated using LYSO-ID® Red dye. (A) Untreated cells (B) Chlorpromazine, 28 µM (C) Verapamil, 200 µM. These two compounds are cationic and amphiphilic, and known to induce abnormal accumulation of phospholipids within lysosomes, resulting in lamellar bodies. Treatment of U-2 OS cells with phospholipidosis-inducing drugs causes an increase in lysosome number and volume, detected as an increase in red fluorescence. Nuclei are counter-stained with Hoechst 33342 dye (blue).

Handling & Storage

Use/Stability	With proper storage, the kit components are stable up to the date noted on the product label. Store kit at -20°C in a non-frost free freezer, or -80°C for longer term storage.
Handling	Protect from light. Avoid freeze/thaw cycles.
Long Term Storage	-80°C
Shipping	Dry Ice

Regulatory Status

RUO - Research Use Only

Product Details

Application	Fluorescence microscopy, Fluorescent detection
Application Notes	96-well cell-based assay. Provides a rapid and quantitative approach for determining drug- or toxic agent-induced lysosome and lysosome-like organelle perturbations in live cells.
Contents	**10x dual color detection reagent**: 2 x 1ml **Detection buffer**: 20ml **Verapamil control**: 3µmoles **10x assay buffer**: 15ml
Quality Control	A sample from each lot of LYSO-ID® Red cytotoxicity kit (GFP-CERTIFIED®) is used to stain epithelial cell line, MDCK, using the procedures described in the user manual. The Z' value, as observed by the fluorescence microplate reader, is > 0.5 for cells treated with 120µM verapamil. The blue nuclear counterstain is at least 70% of the value for the untreated cells.
Quantity	2 x 96-well plates

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