Description: The Resazurin Cell Viability Kit is a fluorescent assay that detects cellular metabolic activity. The blue nonfluorescent resazurin reagent is reduced to highly fluorescent resorufin by dehydrogenase enzymes in metabolically active cells. This conversion only occurs in viable cells and thus, the amount of resorufin produced is proportional to the number of viable cells in the sample. The resorufin formed in the assay can be quantified by measuring the relative fluorescence units (RFU) using a fluorometer (Ex=530-570 nm, Em=590-620 nm).

Background: Cell viability assays are widely used in drug discovery for the study of growth factors, cytokines, and cytotoxic agents. High throughput screening, in both early drug discovery compound screening and subsequent drug safety and toxicity studies, require reliable, sensitive, and simple assays with the ability to analyze a large number of samples. Colorimetric cell viability assays using tetrazolium salt, such as MTT, XTT, and WST-1, were developed based on live cell reduction of tetrazolium salt into highly colored formazan compounds (1,2). Similarly, resazurin (blue and nonfluorescent) can be reduced to resorufin (pink and highly fluorescent) in live cells and is therefore used to assess mammalian cell toxicity, viability, migration, and invasion (3-6). Similar to the XTT Cell Viability Kit #9095, the Resazurin Cell Viability Kit does not require radioactive materials, cell fixation, or cell permeabilization, and cells used in this assay may be used for further analysis.

Specificity/Sensitivity: The Resazurin Cell Viability Kit detects resorufin produced from resazurin conversion by metabolic enzymes in live cells. This kit is expected to work in most cell lines. For most experiments, 0.02-2x10^5 cells/well should be sufficient, but this can vary depending on the cell type and incubation time. For best results, a cell number titration and incubation time course (as shown in Figure 1) is recommended.

Note: Microbial contaminants will reduce resazurin to resorufin, yielding false positive results. Autofluorescent compounds may interfere with this assay.

Background References:

Figure 1. HeLa cells were seeded at varying density in a 96-well plate and incubated overnight. The Resazurin solution (10% of cell culture volume) was added to the plate and relative fluorescent units were measured at 0, 1, 2, 4, and 6 hr.

Figure 2. Jurkat cells were seeded at 1x10^5 cells/well in a 96-well plate and then treated overnight with various concentrations of astemizole or terfenadine. Cytotoxicity was measured using the Resazurin Cell Viability Kit (left), followed by BrdU Cell Proliferation Assay Kit #6813 (right).
Resazurin Cell Viability Assay Protocol

A.

1. Thaw out Resazurin solution (if kept frozen) and warm it to 37°C to ensure all components are completely in solution.
2. Plate cells in 96-well plate (black plate with clear bottom). Typical seed cell number is 0.02-2x10^5 cells/well depending on cell growth rate. Cell number titration is recommended to determine the optimal cell seeding density.
3. Incubate cells with compound of interest for desired period of time (1-72 hr). Make sure all the wells contain the same volume of medium.
4. Add Resazurin solution to plate (10% of the initial volume in the well). For example, for plates containing 100 μl medium/well, add 10 μl resazurin solution to each well.
5. Incubate the plate for 1-6 hr in standard culture conditions. Incubation time depends on cell type and cell number. The plate can be read multiple times to determine the optimal time point.
6. Measure the relative fluorescent units (RFU) using a plate reader: Ex=530-570 nm, Em=590-620 nm.
1. PRODUCT AND COMPANY IDENTIFICATION

Product Name: Resazurin Cell Viability Kit Reagent

Product Number: 11884

Identified Uses: For Research Use Only (RUO). Not intended for use in humans or animals. Not intended for therapeutic or diagnostic procedures.

Manufacturer/Supplier: Cell Signaling Technology, Inc.

2. HAZARDS IDENTIFICATION

Emergency Overview
OSHA Hazards: No known OSHA hazards.

GHS Classification: Non-hazardous substance according to GHS.

Physical State: Liquid

Potential Health Effects
Acute Toxicity
Skin: No information available
Inhalation: No information available
Ingestion: No information available

3. COMPOSITION/INFORMATION ON INGREDIENTS

No ingredients are hazardous according to OSHA, GHS criteria.

5. FIRE FIGHTING MEASURES

Flash Point: No data available
Suitable Extinguishing Media: Use dry chemical, water spray, alcohol-resistant foam or carbon dioxide.
Special exposure hazards: No information available
Special protection for fire fighters: Wear self-contained breathing apparatus and protective unit.

6. ACCIDENTAL RELEASE MEASURES

Personal precautions: Avoid contact with skin, eyes, clothing. Avoid creating dusts during handling. Prevent product from entering drains. Prevent further spillage if it is to do so.

7. HANDLING AND STORAGE

Safe handling advice: Ensure adequate ventilation. Handle in accordance with good industrial hygiene and safety practices.

Storage conditions: Keep container tightly closed in cool dry place.

Incompatibilities: No data available

Specific end uses: No data available

8. EXPOSURE CONTROLS/PERSONAL PROTECTION

Eye Protection: Safety glasses tested and approved under appropriate government standards such as NIOSH, CEN).
Skin and body protection: Wear suitable protective clothing, protective shoes or boots.
Hand protection: Wear compatible chemical resistant gloves. Dispose of gloves in accordance with applicable laws and good laboratory practices.

9. PHYSICAL AND CHEMICAL PROPERTIES

Information on basic physical and chemical properties

Physical State: Liquid
Color: No data available
Odor: No data available
pH: No data available
Boiling point: No data available
Melting point: No data available
Decomposition temperature: No data available

10. STABILITY AND REACTIVITY

Stable under normal conditions.

11. TOXICOLOGICAL INFORMATION

Potential Health Effects
Inhalation: No information available
Ingestion: No information available
Skin Contact: No information available
Eyes: No information available

Acute Toxicity
To the best of our knowledge, the chemical, physical and toxicological properties have not been fully investigated.

Route of Exposure
Inhalation: No data available
Skin: No data available
Eye: No data available
Ingestion: No data available

12. ECOLOGICAL INFORMATION

Bioaccumulation: No data available
Decomposition temperature: No data available
Maximum容许: No data available

13. DISPOSAL CONSIDERATIONS

Waste Disposal Methods: Dispose of in accordance with all applicable environmental laws and regulations.

14. TRANSPORT INFORMATION

Mode of Transport: Not regulated
DOT: Not regulated
 IMDG: Not regulated

15. REGULATORY INFORMATION

OSHA Hazards: No known OSHA hazards

GHS Classification: Not a dangerous substance according to GHS.

U.S. Federal Regulations
SARA 302: No chemicals in this material are subject to reporting requirements established by SARA Title III, Section 302
SARA 311/312: No SARA hazards
SARA 313: Section 313 of Title III of the Superfund Amendments and Reauthorization Act of 1986 (SARA). This product does not contain any chemicals that exceed the reporting levels of the Act and Title IV of the Code of Federal Regulations, Part 372.

End of Material Safety Data Sheet