Cell Viability Assay

Cell vitality refers to the proportion of healthy, active cells within a cell population. The vitality of cells can be assessed by examining ongoing metabolic activities and enzyme activities, among other cellular physiological functions. Abcam offers various reagents for testing cell vitality, including CCK-8, Resazurin, TMRE, Calcein violet, and ATP luminescence.

Cell reducing Ability analysis

Dyes reduced by cellular enzymes

By detecting the ability of cellular enzymes to reduce dyes, the viability of cells can be analyzed. Commonly used cell viability detection dyes include Tetrazolium dye and Resazurin

Tetrazolium salt dyes can penetrate into living cells and are reduced to colored formazan (Formazan) by dehydrogenase in living cells. However, dead cells cannot reduce the dye to formazan due to lack of enzymes, so formazan is The amount of phosphorus formed will be proportional to the number of living cells and reducing ability. There are many different tetrazolium salt derivatives on the market. The color of the formazan formed by each derivative will be different, and the solubility will also be different.

Please see the following table for details:

Tetrazolium Salt	MTT	XTT	MTS	WST-1	WST-8 / CCK-8
Narrate	Original tetrazolium assay; still very popular. Only tetrazolium assay that needs a wash / solubilization step.	The first improved water-soluble tetrazolium salt, which is less stable and difficult to store	Water-soluble tetrazolium salt, Better stability than XTT	WSTs is a series of tetrazolium salt that are a newer generation than WST-1 and have higher water solubility.	The tetrazolium series has the best effect and has better sensitivity than MTT, XTT and MTS.It has high water solubility, high sensitivity and low toxicity.
Principle	Enters the cell and is directly reduced inside the cell	Reduced outside the cell with the help of electron carriers	Reduced outside the cell with the help of electron carriers	Reduced outside the cell with the help of electron carriers	Reduced outside the cell with the help of electron carriers
Formazan color	Purple	Orange	brownish yellow	Orange	Orange
Water soluble	Insoluble in water, Need to be dissolved with DMSO	+	+++	+++	+++++
Reaction time	+++++	+++	+++	+	+
Cytotoxicity	+++++ Destroy cells, cannot be measured repeatedly	++ Low cytotoxicity, repeatable experiments	++ Low cytotoxicity, repeatable experiments	+ Low cytotoxicity, repeatable experiments	+ Low cytotoxicity, repeatable experiments
Sensitivity	+	+++	+++	+++	+++++
Stability	+++	+	+	+++	+++++
Detection wavelength / Instrument	590 nm / Visible light analyzer	450 nm / Visible light analyzer	490 nm / Visible light analyzer	440 nm / Visible light analyzer	460 nm / Visible light analyzer
Catalog No.	ab211091	ab232856	ab197010	ab65473	ab228554

Highly Recommended Products

The best performance among the tetrazolium salt

Cell Counting Kit 8 (WST-8 / CCK8) (ab228554)

- **High solubility** — The product after the CCK-8 reaction has high solubility and does not need to be added DMSO like the MTT test.
- **High Sensitivity** — The sensitivity is higher than MTT, XTT, MTS and WST-1, and the linear range is wide.
- **Low cytotoxicity** — CCK-8has extremely low toxicity to cells, and the cells that have been tested can be used for other experiments.
- **Simple and fast** — The CCK-8 reagent can be used immediately without thawing or additional preparation steps. The product has high solubility, and the absorbance value can be directly detected after the reaction without additional steps to dissolve the product.
- **Suitable for Long-term culture** — Suitable for long-term culture CCK-8 has high stability and extremely low cytotoxicity, so it is suitable for experiments requiring longer culture time (for example: 24~48 hours)

- Resorufin dye can penetrate into living cells, accept electrons in the mitochondrial respiratory electron transport chain, and then be reduced to pink color, and produce a fluorescent product_(Resorufin), The absorbance value or fluorescence intensity of fluorine is directly proportional to the number of cells and reducing ability. Therefore, cell activity can be evaluated through visible light absorbance detection or fluorescence detection.

Method	Instrument	Description	Item number
Resazurin	**Visible light** **Fluorescence analyzers** **Microscopes** **Flow cytometers**	▸ Fluorescent light Ex/Em: 535–560 /560–615 nm ▸ Visible light 570 nm	**ab129732**

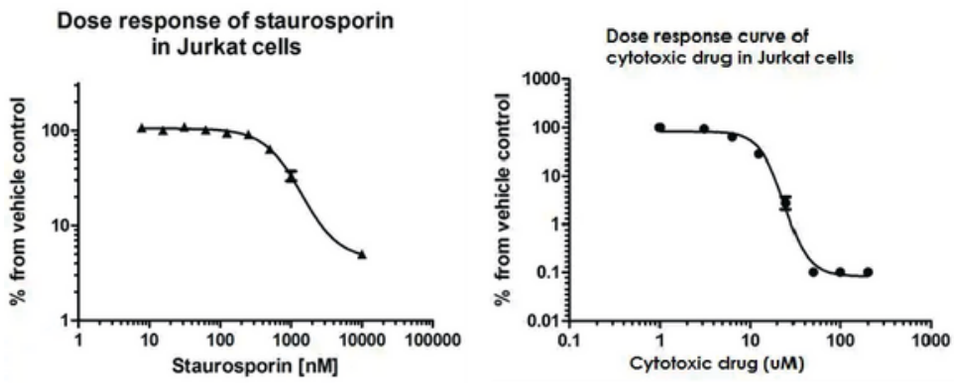

Figure 1. Jurkat cells treated with idarubicin (A) or staurosporin (B) were analyzed with Resazurin assay ab129732.

Mitochondrial membrane potential dependent dyes

Mitochondrial membrane potential, whether high or low, can indicate the cellular vitality status and can be utilized to identify live cells through the accumulation of specific dyes within the mitochondria.
Conversely, the loss of mitochondrial membrane potential and the inability of dyes to stain are often employed to detect cell apoptosis.

Method	Instrument	Description	Item number
TMRE/TMRM	FA、FM 、FC	▸ Most Popular Mitochondrial Membrane Stain ▸ Detect fluorescence Ex/Em 549/575 nm	ab113852
JC-1	FA 、FM 、FC	▸ JC-1 will aggregate to form color when the mitochondrial membrane potential is changing High=red , Low=green ▸ Detect fluorescence Ex/Em 530/530-570 nm	ab113850
JC-10	FA 、FM 、FC	▸ Principle same as JC-1 but the water solubility is higher ▸ Detect fluorescence Ex/Em 590/520-570 nm	ab112134 ab112133
Rhodamine 123	FA 、FM 、FC	▸ Detect fluorescence Ex/Em 507/529 nm	ab275545
MitoNIR	FA	▸ Detect fluorescence Ex/Em 635/660 nm	ab112149 ab112150
MitoOrange	FA	▸ Detect fluorescence Ex/Em 540/590 nm	ab138898 ab138899

FA=Fluorescence Analyzer
FM=Fluorescence Microscopy
FC=Flow Cytometry
LM=Luminometer

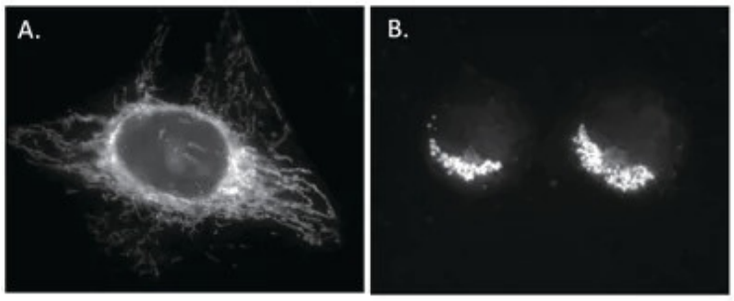

Figure 2. Cell staining with TMRE kit ab113852.
A: Healthy HeLa cells. B: Healthy Jurkat cells.

Cellular esterase cleaved dyes

Evaluated by detecting intracellular esterase activity Cell viability. Calcein and similar hydrophobic dyes diffuse into cells and are cleaved by intracellular esterases in live cells. The hydrophilic fluorescent product is retained within the cell.Detecting the production of fluorescent products can evaluate the enzyme activity of lactonase and reflect the cell activity state.

Method	Instrument	Description	Item number
Calcein AM	FA、FM、FC	▸ Ex/Em 495/515 nm	ab141420
Calcein violet AM	FA、FM、FC	▸ Ex/Em 405/460 nm	ab176748
Esterase-cleaved blue	FA	▸ Ex/Em 360/450 nm	ab112120
Esterase-cleaved green	FA、FM	▸ Ex/Em 490/520 nm	ab112122

ATP and ADP assays

ATP is the energy source of living cells, ATP level can reflect mitochondrial activity and cell activity status. Common ATP detection method is to first degrade the cells to release the ATP in the cells. The released ATP reacts with ATP-dependent (Luciferase) to produce light. ATP detection uses ATP phosphorylation activity to detect ATP levels, allowing ATP to phosphorylate oil (or other substrates). The phosphorylated products can be detected by VIsible light or fluorescence.

Method	Instrument	Description	Item number
Luminescence ATP assay	LM	▸ Luminometric plate reader.	ab113849
Luminescence ADP/ATP	LM	After ATP analysis, ▸ ADP is converted to ATP for detection	ab65313
ATP phosphorylation	FA	▸ Not as sensitive as luminescence assays. ▸ more sensitive than colorimetric. Plate reader ▸ Visible light 570 nm Fluorometric Ex/Em 570 nm	ab83355

Oxygen consumption and glycolysis assays

The rate of oxygen consumption indicates the level of cellular metabolic activity. Analysis of intracellular oxygen levels and glycolysis activity allow deeper investigation.

Method	Instrument	Description	Item number
Extracellular oxygen consumption	FA	▸ Dye signal Increases as respiration ▸ Ex/Em 380/650 nm	ab113849
Intracellular oxygen levels	FA	▸ Dye signal is quenched by intracellular oxygen ▸ Ex/Em 340/642 nm	ab65313
Glycolysis activity	FA	▸ Lactate production causes extracellular acidification and increased dye fluorescence ▸ Ex/Em 340-380/615 nm	ab83355

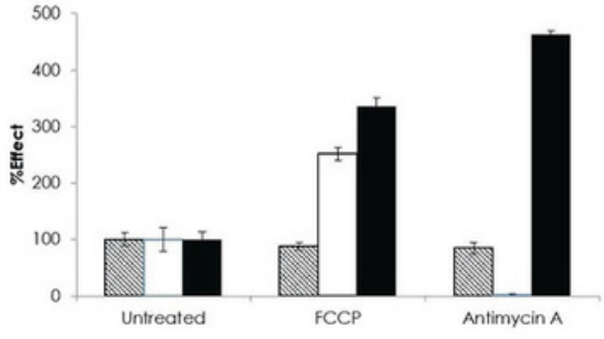

Figure 3. HepG2 cells treated with antimycin A and FCCP and tested with assays for ATP (gray, ab113849, oxygen consumption (white, ab197243) and glycolysis (black, ab19724)

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