Catalog Number	Unit Size
TBS2022-500	500 Assay
TBS2022-1K	1000 Assay
TBS2022-5K	5000 Assay

DESCRIPTION

TribioScience's Cell Counting Kit-8 (CCK-8) is a colorimetric assay to quantify the viable cells, consisting of the tetrazolium salt and the electron mediator 1-mthoxyPMS. The working principle is based on the dehydrogenase activity detection. The generated NADH from the reaction of dehydrogenase enzymes and the substrates oxidizes the 1-methoxyPM. The reduced 1-methoxyPMS further oxidizes the tetrazolium salt (WST-8) in CCK-8 assay kit, forming the yellow formazan dye. The dye color can then be quantified by measuring the absorbance at 460 nm wavelength so that cell number is known. The assay can be used for the measurement of cell proliferation in response to growth factors, cytokines, mitogens, and nutrients, etc. It can also be used for the analysis of cytotoxic compounds like anticancer drugs and many other toxic agents and pharmaceutical compounds.

FEATURES

- **Simplicity:** One-bottle, ready-to-use solution. No harvesting, no washing, and no solubilization steps.
- **Safety**: No organic solvents or isotopes required.
- **Accuracy**: Absorbance measurement is proportional to the number of cells in each well.
- **Sensitivity:** More sensitive than MTT, XTT, MTS or WST-1.

Kit Components and Storage for 500 tests

Name	500 Assays	1000Assay	5000Assays
CCK-8 Reagent	5 mL	10 mL	50 mL

Storage Condition: 2-8 °C
Shelf-life: 1 years

APPLICATIONS

- Cell proliferation
- Drug screening
- Growth factors
- Cytotoxicity
- Apoptosis

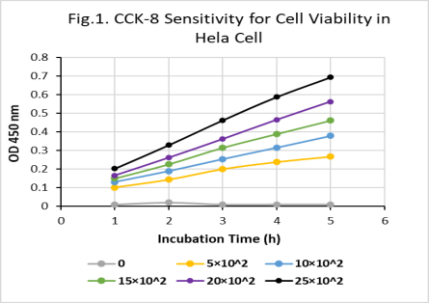
DIRECTIONS FOR USE

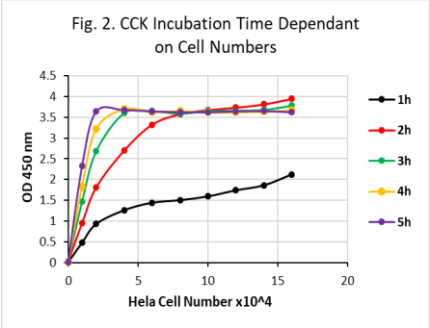
Cell Proliferation Assay

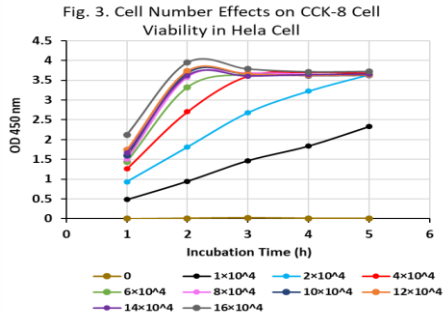
1. Inoculate cell suspension (100 µl/well) in a 96-well plate. Also prepare wells that contain known numbers of viable cells (to create a calibration curve in step 5). Pre-incubate the plate in a humidified incubator (e.g., at 37 °C, 5% CO2).
2. Thaw the CCK-8 on the bench top or in a water bath at 37 °C if it is frozen. *Note: It takes about 30 minutes on the bench top at 25 °C or 5 minutes in a water bath at 37 °C.*
3. Add 10 µl of the CCK-8 solution to each well of the plate. *Note: Be careful not to introduce bubbles to the wells, since they interfere with the O.D. reading.*
4. Incubate the plate for 1-4 hours in the incubator.
5. Measure the absorbance at 450 nm using a microplate reader. Prepare a calibration curve using the data obtained from the wells that contain known numbers of viable cells. Note: To measure the absorbance later, add 10 µl of 1% w/v SDS to each well, cover the plate and store it with protection from light at room temperature. No absorbance change should be observed for 48 hours.

Cytotoxicity Assay

1. Dispense 100 µl of cell suspension (5000 cells/ well) in a 96-well plate.
2. Pre-incubate the plate for 24 hours in a humidified incubator (e.g., at 37 °C, 5% CO2).
3. Add 10 µl of various concentrations of toxicant into the culture media in the plate.
4. Incubate the plate for an appropriate length of time (e.g., 6, 12, 24 or 48 hours) in the incubator.
5. Thaw the CCK-8 on the bench top or in a water bath at 37 °C if it is frozen. Note: It takes about 30 minutes on the bench top at 25 °C or 5 minutes in the water bath at 37 °C.
6. Add 10 µl of CCK-8 solution to each well of the plate. Be careful not to introduce bubbles to the wells, since they interfere with the O.D. reading.
7. Incubate the plate for 1-4 hours in the incubator. Measure the absorbance at 450 nm using a microplate reader. *Note: To measure the absorbance later, add 10 µL of 1% w/v SDS to each well, cover the plate and store it with protection from light at room temperature. No absorbance change should be observed for 48 hours.*







RELATED PRODUCTS

Resazurin Cell Viability (TBS2001)
LDH Cytotoxicity Assay (TBS2002)
MTT Cell Viability Assay (TBS 2003)
MTS Cell Viability Assay (TBS2004)
Catalase Assay (TBS2006)
ATP Colorimetric/ Fluorometric Assay (TBS 2010)
ADP Colorimetric/Fluorometric Assay (TBS3020)
XTT Cell Viability Assay (TBS2021)
Caspase-3 Colorimetric Assay (TBS2030)
BrdU Cell Proliferation Colorimetric Assay (TBS2086)
Cytochrome c Reductase Activity Assay (TBS2116)
AOPI Viability Assay for Flow Cytometry (TBS2069

Comparison between CCK-8 and other methods

Properties	CCK8	MTT	XTT	WST-1
Solubility	+	-	+	+
Forms	1-bottle solution	Powder	2-bottle solution	1-bottle solution
Preparation	Ready to use	Dissolve before	Mix before use	Ready to use
Sensitivity	+++	+	++	++
Detection	+++	+	++	++
Wavelength	430~490 nm	560~600 nm	420~480 nm	420~480 nm
Toxicity	-	+	-	-
Stability	++	+	-	+
Convenience	+++	+	++	++

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