Datasheet for ABIN1000242

Cell Viability Assay Kits

16 Publications

Go to Product page

Overview

Quantity:	500 tests
Application:	Cellular Assay (CA)

Product Details

Characteristics:	Safe. Non-radioactive assay (cf. 3 H-thymidine incorporation assay). Sensitive and accurate. As low as 950 cells can be accurately quantified. Fast. High-throughput assay using 96-well plates allows simultaneous processing tens of thousands of samples per day. Homogeneous and convenient. Mix-incubate-measure type assay. No wash and reagent transfer steps are involved. Robust and amenable to HTS. Z' factors of 0.5 and above are observed. Can be readily automated with HTS liquid handling systems.

Target Details

Background:	Colorimetric (570nm) assay for cell viability, proliferation, cytotoxcity, HTS for anticancer agents. The study of cell proliferation and cell viability requires the accurate quantification of the number of viable cells in a cell culture. Therefore, assays for calculating cell viability are necessary for optimizing cell culture conditions, evaluating cell growth factors and nutrients, discovering novel antibiotics and anti-cancer drugs, evaluating toxic effects of environmental pollutants and cell mediated toxicity and studying programmed cell death (apoptosis). The CellQuanti-MTT TM assay kit provides a convenient, sensitive, quantitative and reliable assay

Target Details

for determining the number of viable cells in a given culture. This homogeneous colorimetric assay is based on the conversion of a tetrazolium salt MTT, a pale yellow substrate, to formazan, a purple dye. This cellular reduction reaction involves the pyridine nucleotide cofactors NADH/NADPH and is only catalyzed by living cells. The formazan product has a low aqueous solubility and is present as purple crystals. Dissolving the resulting formazan with a solubilization buffer permits the convenient quantification of product formation. The intensity of the product color, measured at 550 - 620 nm, is directly proportional to the number of living cells in the culture. Reagents in the kit have been carefully formulated and optimized for sensitivity, assay robustness and automation.

Application Details

Application Notes:	Cell Proliferation: effects of cytokines, growth factor, nutrients. Cytotoxicity and Apoptosis: evaluation of toxic compounds, anti-cancer antibodies, toxins, environmental pollutants etc. Drug Discovery: high-throughput screen for toxic and anticancer drugs.
Protocol:	The assay is based on the conversion of MTT to purple formazan by metabolically active cells. For most cells, this reducing reaction takes 3 to 5 hours. The produced formazan is solubilized and the resulting colored solution is quantified with a microplate reader. Although most culture media contain phenol red, phenol red does not interfere with the assay. All data in Technical Notes were obtained in culture media containing phenol red. Procedure using 96-well plate: 1. Plate and culture cells (80 µL per well) in a clear bottom 96-well tissue culture plates. Typical culture medium contains DMEM, 10% fetal bovine serum, antibiotics (penicillin/streptomycin, gentamycin, etc), amino acids and other nutrients. Assays can be performed on either adherent cells or cells in suspension. The number of cells can vary from 1,000 to 80,000 per well. The volume can vary from 50 to 150 µL, although 80 µL is used in this example. In addition to the test samples, one must include control wells of culture medium containing no cells or cells treated with a toxic reagent such as 0.1% saponin. 2. Add test compounds and controls and incubate cells for the desired period of time (typically overnight). It is recommended that assays be run in duplicate or triplicate. A volume of 20 µL in phosphate buffered saline (PBS) or culture medium is recommended for the test compounds and controls. The Control reagent can be conveniently reconstituted with 5 mL PBS (1% saponin). 3. Reconstitute the Reagent with Assay Buffer. First equilibrate the Reagent and Assay Buffer to

Application Details

	room temperature. Then simply combine Assay Buffer and the Reagent by pipetting a small volume (e.g. 1 mL) buffer to the Reagent tube. Vortex briefly and pipet the reconstituted solution to the Assay Buffer bottle. Repeat this step to transfer all Reagent to the Assay Buffer bottle. Mix by inversion until the Reagent is thoroughly dissolved. After this is done, mark the bottle label as Reconstituted Reagent. Note: Fresh reconstitution is recommended although the reconstituted reagent is stable for up to 4 weeks when stored at -20 °C. 4. Add 15 µL (per 80 µL cell culture) of reagent per well and incubate for 4 hours at 37°C. The volume of the reagent should be adjusted depending on the volume of cell culture. 5. Add 100 µL of the Solubilization Solution. Mix gently on an orbital shaker for one hour at room temperature. The volume of the Solubilization Solution should be adjusted depending on the volume of cell culture. If precipitation occurs in the Solubilization Solution, place the bottle in a warm water bath or at 37°C and shake to dissolve precipitates. 6. Measure OD570nm for each well on an absorbance plate reader. Maximum absorbance of the formazan dye lies between 560 and 590 nm. If desired, the OD measurement can be performed the following day. In this case, it is recommended to seal the plate to minimize evaporation.
Restrictions:	For Research Use only

Handling

Storage:	4 °C

Publications

Product cited in:	Harbison, Ryan, Wilkins, Schroeder, Loucks, Bouchard, Linseman: "Calpain plays a central role in 1-methyl-4-phenylpyridinium (MPP+)-induced neurotoxicity in cerebellar granule neurons." in: **Neurotoxicity research**, Vol. 19, Issue 3, pp. 374-88, (2011) (PubMed). Kalaivani, Rajasekaran, Suthindhiran, Mathew: "Free Radical Scavenging, Cytotoxic and Hemolytic Activities from Leaves of Acacia nilotica (L.) Wild. ex. Delile subsp. indica (Benth.) Brenan." in: **Evidence-based complementary and alternative medicine : eCAM**, Vol. 2011, pp. 274741, (2011) (PubMed). Cheng, Feng, Figueiredo, Zhang, Nelson, Marigo, Beck: "Transient receptor potential melastatin type 7 channel is critical for the survival of bone marrow derived mesenchymal stem cells." in: **Stem cells and development**, Vol. 19, Issue 9, pp. 1393-403, (2010) (PubMed).

Poh, Shi, Lim, Neoh, Wang: "The effect of VEGF functionalization of titanium on endothelial cells in vitro." in: **Biomaterials**, Vol. 31, Issue 7, pp. 1578-85, (2010) (PubMed).

André, Rassokhin, Bowman, Pakhomov: "Gadolinium blocks membrane permeabilization induced by nanosecond electric pulses and reduces cell death." in: **Bioelectrochemistry (Amsterdam, Netherlands)**, Vol. 79, Issue 1, pp. 95-100, (2010) (PubMed).

There are more publications referencing this product on: Product page