

CellTiter 96[®] AQ_{ueous} One Solution Cell Proliferation Assay

The standard in colorimetric cell proliferation and cytotoxicity assays

The CellTiter 96[®] AQ_{ueous} One Solution Cell Proliferation Assay^(a) is a colorimetric method for determining the number of viable cells in proliferation, cytotoxicity or chemosensitivity assays. The CellTiter 96[®] AQ_{ueous} One Solution Reagent is a stable mixture containing a novel tetrazolium compound (3-(4,5-dimethylthiazol-2-yl)-5-(3-carboxymethoxyphenyl)-2-(4-sulfophenyl)-2H-tetrazolium, inner salt: MTS^(a)) and an electron coupling reagent (phenazine ethosulfate; PES). The concentrations of MTS and electron coupling reagent have been optimized for use with a variety of anchorage-dependent and suspension cell types, including human lymphocytes. The CellTiter 96[®] AQ_{ueous} One Solution Cell Proliferation Assay can be used in variety of applications including cell proliferation, cytotoxicity, chemosensitivity, chemotaxis, apoptosis, and cell attachment studies for screening antiviral compounds.

Assays are performed by adding a small amount of the CellTiter 96[®] AQ_{ueous} One Solution Reagent directly to culture wells, incubating for 1–4 hours, then recording absorbance at 490nm with a spectrophotometric plate reader (Figure 1). MTS tetrazolium is bio-reduced by cells into a colored formazan product that is soluble in tissue culture medium (Figure 2). The quantity of formazan product formed is directly proportional to the number of living cells in culture (Figure 3).

The CellTiter 96[®] AQ_{ueous} One Solution Reagent can be directly substituted in many applications where proliferation is measured using [³H]thymidine incorporation. A comparison of results obtained using the two assay procedures is shown in Figure 4, demonstrating that the critical parameter of ED₅₀ (the quantity of factor resulting in half the maximal response) is similar in both assays.

Visit Promega's web page at: www.promega.com/cellprolif/ to access the Cell Viability Assistant, a database of citations reporting the use of Promega cell viability assays with a variety of cell types in numerous applications.

Benefits:

- **Simple:** "Add-incubate-measure" format (single-step reagent addition) enables design of homogeneous high-throughput screening assays.
- **Convenient:** Supplied as a single solution, filter sterilized and ready to add to assay plates (unlike MTT assays).
- **Fast:** Perform the assay in 96-well plates with no washing or cell harvesting. No solubilization steps required (unlike MTT assays).
- **Non-Radioactive:** Requires no scintillation cocktail or radioactive waste disposal (unlike [³H]thymidine incorporation assays).
- **Flexible:** Plates can be read and returned to incubator for further color development (unlike MTT assays).
- **Safe:** Requires no volatile organic solvent to solubilize the formazan product (unlike MTT assays).

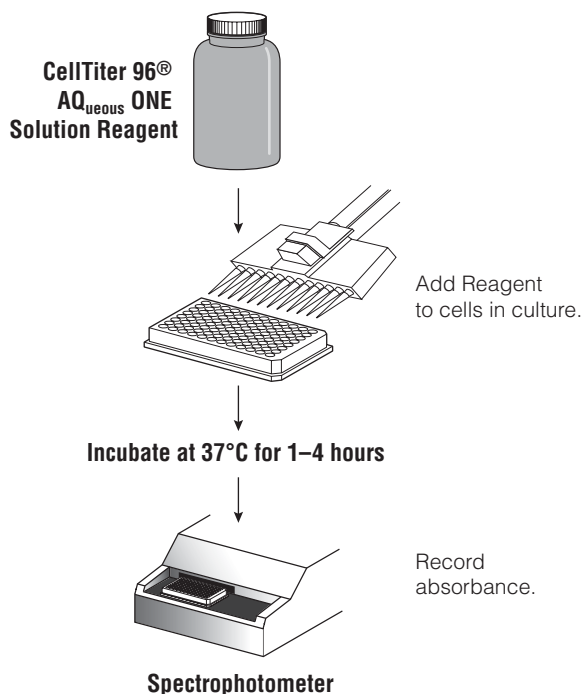


Figure 1. Schematic diagram of the CellTiter 96[®] AQ_{ueous} One Solution Assay.

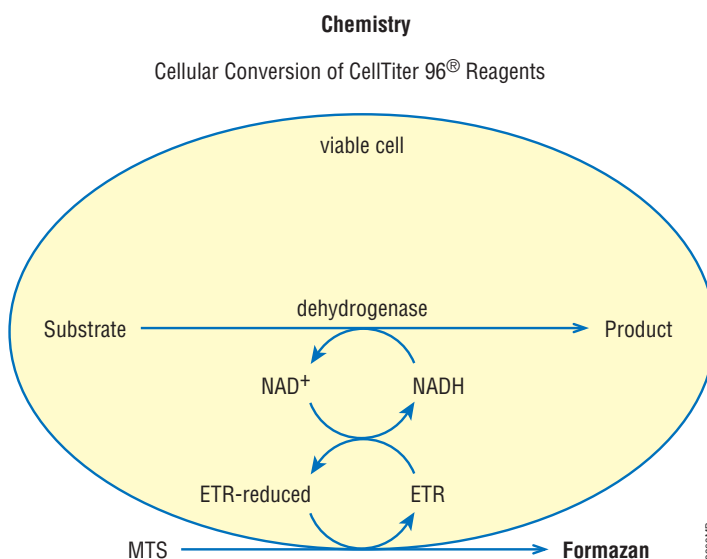


Figure 2. Schematic diagram of bio-reduction of tetrazolium compounds by viable cells.

Ordering Information

Product	Size	Cat.#
CellTiter 96 [®] AQueous One Solution Cell Proliferation Assay ^(a)	200 assays	G3582
	1,000 assays	G3580
	5,000 assays	G3581

For Laboratory Use.

Literature

Protocol	Part#
Technical Bulletin	TB245

Related Products

Cell Viability Assays	Cat.#
CellTiter 96 [®] AQueous MTS Reagent Powder ^(a)	G1112
CellTiter-Glo [®] Luminescent Cell Viability Assay ^(b,c) (ATP detection, luminescence)	G7570
CellTiter-Blue [™] Cell Viability Assay (Resazurin reduction, fluorescence)	G8080
CellTiter 96 [®] Non-radioactive Cell Proliferation Assay (MTT reduction, absorbance)	G4000
CytoTox-ONE [™] Homogeneous Membrane Integrity Assay ^(d) (LDH release, fluorescence)	G7890
CytoTox 96 [®] Non-Radioactive Cytotoxicity Assay* (LDH release, absorbance)	G1780

Apoptosis Assays	Cat.#
Caspase-Glo [™] 3/7 Assay ^{*(b,c)} (luminescence)	G8090
Caspase-Glo [™] 8 Assay ^{*(b,c)} (luminescence)	G8200
Caspase-Glo [™] 9 Assay ^{*(b,c)} (luminescence)	G8210
Apo-ONE [®] Homogeneous Caspase-3/7 Assay (fluorescence)	G7792

* For Laboratory Use.

^(a) The MTS tetrazolium compound is the subject of U.S. Pat. No. 5,185,450 assigned to the University of South Florida and is licensed exclusively to Promega Corporation.

^(b) U.S. Pat. No.6,602,677, Australian Pat. No. 754312 and other patents pending.

^(c) The method of recombinant expression of *Coleoptera* luciferase is covered by U.S. Pat. Nos. 5,583,024, 5,674,713 and 5,700,673.

^(d) Patent Pending.

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Absorbance is Directly Proportional to Number of Cells

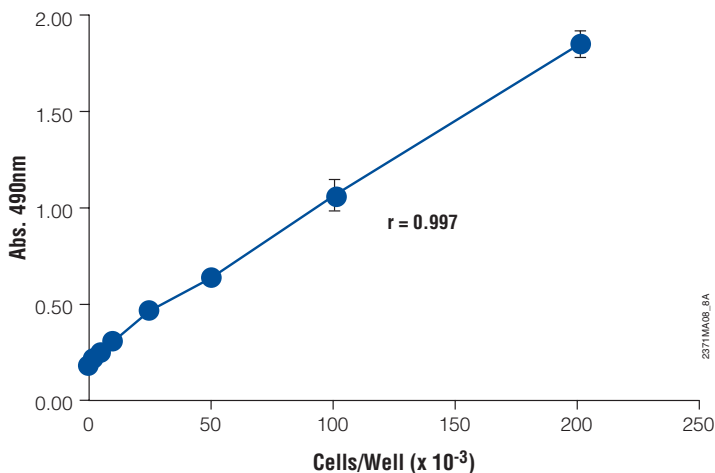


Figure 3. Effect of cell number on absorbance at 490nm measured using the CellTiter 96[®] AQueous One Solution Assay and B9 hybridoma cells.

Comparison of MTS and [³H]thymidine Assays Proliferation of HT-2 Cells Stimulated with GM-CSF

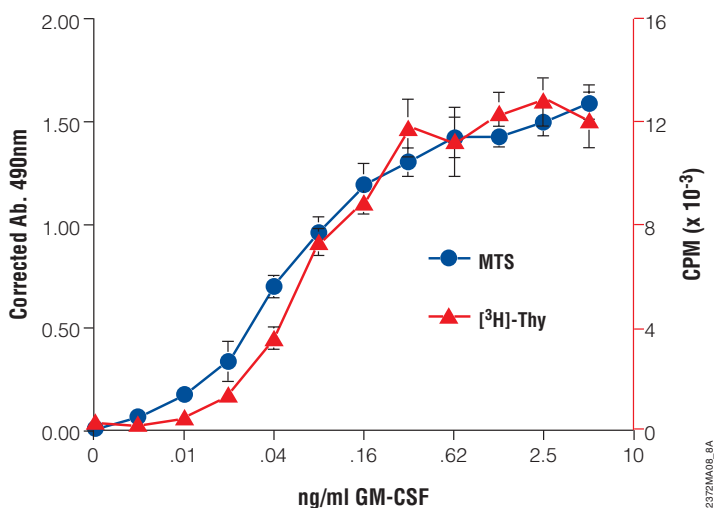


Figure 4. Measurement of GM-CSF-stimulated proliferation in HT-2 cells using the CellTiter 96[®] AQueous Cell Proliferation Assay and a [³H]thymidine incorporation assay. Similar results were obtained with both assays.



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