

Assay Solutions

Easy-to-use, Relevant Assays to Assess Cell Health and Function



Assay Solutions

Lonza offers a range of biochemical, luminescent and cell-based assays that provide a wealth of information relating to your cells; from their state of health to the finest detail of their intracellular signaling mechanisms. These assays can become key tools in your pre-clinical drug discovery processes, from monitoring the quality of the cells you are using to target identification and validation, compound hit and safety screening.

The ViaLight™ Plus Cell Viability Assay is designed to deliver a high, stable luminescent signal for an extended period of time for greater experimental design flexibility. The ToxiLight™ BioAssay Kit is a bioluminescent, non-destructive cytolysis assay kit designed to measure the release of the enzyme adenylate kinase (AK) from damaged cells. For fat and bone-like cells, we provide a distinct range of assay tools to measure lipid accumulation, lipolysis, and bone resorption or mineralization.

Adipocytes, MSCs, and ADSCs can have lipid metabolism measured with AdipoRed™ Adipogenesis Assay Reagent and AdipoLyze™ Lipolysis Assay. Bone cells like osteoclasts and osteoblasts, and even differentiated MSCs and ADSCs can have bone remodeling measured with OsteoAssay™ Human Bone Plate, OsteoLyse™ Bone Resorption Assay Kit and OsteoImage™ Bone Mineralization Assay. All of these assays will support both primary cells and related cell lines.

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ViaLight™ – Measure Cell Proliferation and Cytotoxicity

 $\textbf{ToxiLight}^{\scriptscriptstyle{\text{TM}}}- \textbf{Non-Destructively Check Cytotoxic Compound Effects}$

 $\textbf{PDELight}^{\scriptscriptstyle{\intercal}}- \text{HTS Phosphodiesterase Assay}$

PPiLight™ - Inorganic Pyrophosphate Assay

Cell Type and Enzyme Specific Assays

AdipoRed™ - Adipogenesis Assay Reagent

AdipoLyze™ – Lipolysis Assay

OsteoAssay™ — Human Bone Plate

OsteoLyse™ - Bone Resorption Assay Kit (Human Collagen)

Osteolmage™ - Bone Mineralization Assay



Luminescent Bioassays

ViaLight™ Plus

Measure Cell Proliferation and Cytotoxicity

- Uses bioluminescent detection of cellular ATP as a measure of viability
- Designed to deliver a high, stable luminescent signal for an extended period of time for greater experimental design flexibility

Benefits

Fast: Results from a 96-well plate can be processed and analyzed in <15 minutes

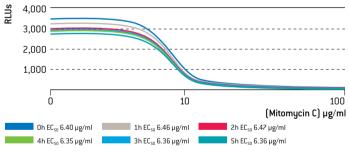
Sensitive: Detect as few as ten cells allowing lower seeding densities and more assays

Convenient: Simple, no-shake protocol for easy and scalable automation — add two reagents directly to your culture well and read luminescence

Flexible: Dynamic range of five decades with both adherent or suspension cell cultures, can be run on a variety of luminometers or scintillation counters

Safe: No radioactivity or toxic components required

EC 50 Data Generated Using ViaLight Plus Shows Consistency Over Time



HepG2 cells were incubated with the alkylating agent Mitomycin C for 48 hours and the assayed using ViaLight™ Plus. The experimental values are the mean of eight replicant samples read every hour over a 5 hour period. The EC values remain consistent over the 5 hour read period.

Ordering Information

Cat. No.	Description	Size
LT07-221	ViaLight™ Plus Cell Proliferation and Cytotoxity BioAssay Kit	500 tests
LT07-121	ViaLight™ Plus Cell Proliferation and Cytotoxity BioAssay Kit	1,000 tests
LT07-321	ViaLight™ Plus Cell Proliferation and Cytotoxity BioAssay Kit	10,000 tests
LT17-221	ViaLight™ Plus Cell Proliferation and Cytotoxity BioAssay Kit	500 tests (with 5 white TC plates)
LT17-517	ViaLight™ 100% Lysis Control Set (sold separately)	10 ml (200 tests)

www.lonza.com/vialight

ToxiLight™

Non-Destructive Cytotoxicity Bioassay Kit

- Designed to measure the leakage of adenylate kinase (AK) from damaged cells
- AK catalyzes the conversion of ADP to ATP which is then measured using a bioluminescent reaction.

Benefits

Highly sensitive: As few as 10 cells, due to the cyclic nature of the AK reaction

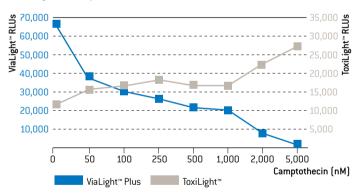
Non-destructive: Eliminating the need to lyse, cytotoxicity can be monitored from a sample of supernatant

Simple: Addition of a single reagent directly to your cells or supernatant

Fast: Results from a 96-well plate can be processed and analyzed in < 10 minutes

Flexible: Supernatants can be frozen with no loss of AK activity (for long-term studies)

Identify Dose-dependent Activities in Cells



Comparison of ViaLight™ Plus and ToxiLight™ Kits using HUVECs dosed with camptothecin. The ATP levels indicated by the ViaLight™ Plus RLUs reduce steadily in a dose-dependent manner. At the lower drug doses, the AK released from the cells is relatively low compared with that of the control, only increasing dramatically at the highest drug doses.

Ordering Information

Cat. No.	Description	Size
LT07-217	ToxiLight™ Non-destrucitve Cytotoxity BioAssay Kit	500 tests
LT07-1117	ToxiLight™ Non-destrucitve Cytotoxity BioAssay Kit	1,000 tests
LT07-117	ToxiLight™ Non-destrucitve Cytotoxity BioAssay Kit	500 tests (with 5 white TC plates)

www.lonza.com/toxilight

PDELight™

High-throughput Screening Phosphodiesterase Assay

- Luminescent assay to identify inhibitors of phosphodiesterase activity
- Luciferase-based system quantifies AMP produced from the hydrolysis of cyclic AMP by phosphodiesterases

Benefits

Simple: Only one reagent to add

Generic: The same assay can be used for all cAMP dependent

phosphodiesterases

Fast: Complete a 384-well plate in less than 3 minutes Versatile: Scalable to 96-, 384-, or 1536-well formats

PPiLight™

Inorganic Pyrophosphate Assay

- Non-radioactive bioluminescent assay for the detection of inorganic pyrophosphate (PPi)
- In the presence of PPi, the conversion of AMP to ATP is catalyzed.
 Luciferase produces light from the newly formed ATP.

Benefits

Fast: Measure enzyme activity via pyrophosphate production

Simple: 2-step luminescent assay; no radioactive substrates, beads,

or antibodies required

Wide detection range: Linear range from 0.02 μM to 10 μM PPi

Sensitive: Down to $0.02\,\mu M$

Versatile: Scalable to 96-, 384-, and 1586-well formats

Cell Function Assays

AdipoRed™ Assay Reagent

Quantify Intracellular Lipid Accumulation

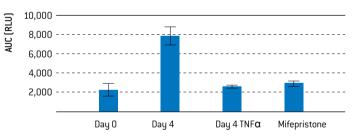
- Assess the effect of compounds on the differentiation of preadipocytes or lipid utilization in mature adipocytes
- AdipoRed™ Assay Reagent specifically partitions into the fat droplets of differentiated adipocytes and fluoresces at 572 nm

Benefits

Convenient: Simply replace cell culture medium with PBS, add AdipoRed™ Reagent and read in a standard fluorimeter Fast: Process an entire 96-well plate in as little as 20 minutes. Much faster and easier than Northern and Western blots Effective: Provides objective high-throughput measurement of the accumulation of intracellular triglycerides with high signal-to-noise ratios

Sensitive: More sensitive than other methods, such as the Oil Red O assay

Inhibition Of Adipocyte Differentiation Assayed With AdipoRed™ Assay Reagent



Poietics[™] Primary Human Preadipocytes were induced to differentiate in the presence of TNFα, Mifepristone or no inhibitor. Lipid accumulation was assayed after 4 days in culture.

Cat. No.	Description	Size
LT07-600	PDELight™ HTS cAMP Phospodiesterase	500 tests
LT07-610	PPiLight™ Inorganic Pryophosphate Assay	500 tests

AdipoLyze™

Lipolysis Detection Assay

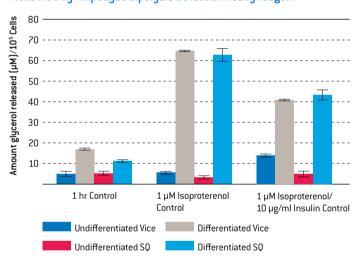
- Fluorescently detect small quantities of glycerol in cells undergoing lipolysis
- Quantitate in vitro lipolysis of adipogenic cell lines as well as primary cells of both subcutaneous and visceral origin

Benefits

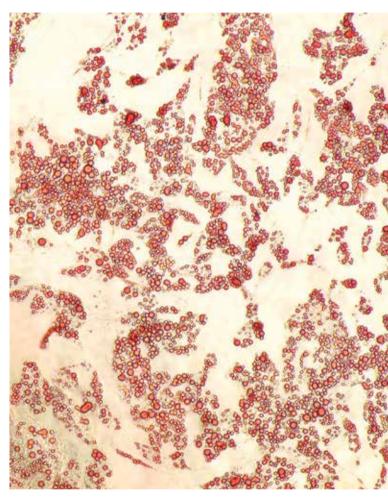
Fast: Completed in < 2 hours

Sensitive: Detects very low levels of glycerol ($0.44\,\mu\text{M}$ or $0.04\,\mu\text{g/ml}$) Versatile: For use in 96-well plate format, but scalable to 384-well

Glycerol Released by Subcutaneous and Visceral Preadipocyte in PGM-2, Measured by AdipoLyze Lipolysis Detection AssayReagent



Poietics™ Human Subcutaneous and Visceral Preadipocytes (undiff & differentiated) treated with isoproterenol alone or with insulin. Lipolysis measured with AdipoLyze™ Assay Kit.



Differentiated visceral adipocytes stained with AdipoRed™ Assay Reagent.

Cat. No.	Description	Size
PT-7009	AdipoRed™ Assay Reagent	5×4.0 ml
00193339	AdipoLyze™ Lipolysis Detection Assay	1×96-wells

OsteoAssay™ Human Bone Plate

Measure Osteoclastic Bone Resorption

- Thin layer of adherent human bone (chips) for the culture of primary human or non-human osteoclasts, osteoclast precursors, and immortalized cell lines
- Measure bone resorption and/or enzyme activity by sampling the cell culture supernatant

Benefits

Convenient: Ready-to-use plates with human bone chips attached to the wells eliminates the need for dentine or animal bone slices

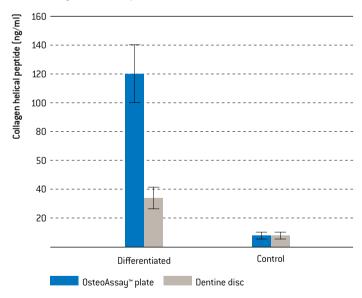
Simple: Cells can be seeded onto the surface of the OsteoAssay™

Plate and used in traditional cell culture protocols

Flexible: Can be used with a variety of cell types and cell-based assays

Novel: Contains real human bone for more biologically relevant results

OsteoAssay™ Plate Is Superior To Dentine Slices



Comparison of primary human osteoclast function (in vitro bone resorption) grown on OsteoAssay $^{\rm m}$ Plate vs. dentine slices.

OsteoLyse™ Assay Kit (Human Collagen)

Measure Bone Resorption in Minutes

- Measure in vitro osteoclast-mediated bone matrix resorption in a high-throughput format
- Kit includes a 96-well cell culture plate coated with europium-labeled human Type I collagen and a bottle of Fluorophore Releasing Agent
- Assay directly measures the release of europium-labeled collagen fragments (resorptive activity) into the osteoclast cell culture supernatant via time resolved fluorescence

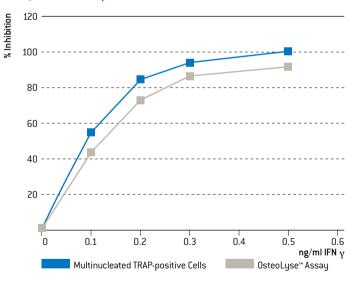
Benefits

Convenient: Human collagen is bound to wells in the plate eliminating the need to purchase bone matrices separately

Easy-to-use: Cells can be seeded onto the surface of the OsteoLyse™ Plate and used in traditional cell culture protocols

Homogeneous: Resorptive activity is easily measured by sampling the cell culture supernatant and counting via time-resolved fluorescence

Comparison of the TRAP Stain and the OsteoLyse™ Assay Kit in an Assay of Interferon y-inhibition of Osteoclast Precursor Differentiation



Poietics™ Human OCP were differentiated on OsteoLyse™ Plate and assessed after 9 days for collagen peptide release using OsteoLyse™ Assay and or TRAP-positive multinucleated cells.

Cat. No.	Description	Size
PA-1500	OsteoLyse™ Assay Kit (HumanCollagen)	96-wells
PA-1000	OsteoAssay™ Human Bone Plate	96-wells

Osteolmage™ Mineralization Assay

Rapid Fluorescent Bone Mineralization Assay

- Quantitate in vitro mineralization by osteogenic stem cells, primary osteoblasts, and osteoblast-like cell lines
- Based on specific binding of the fluorescent Osteolmage™ Staining Reagent to the hydroxyapatite portion of bone-like nodules deposited by cells

Benefits

Novel: Measures hydroxyapatite, similar to real bone **Quick:** Completed in less than 90 minutes with no tedious

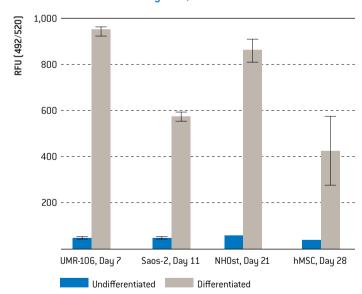
extractions for quantitation

Sensitive: Detects time-dependent increases in mineralization in

differentiating cells

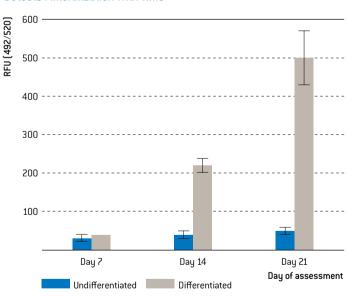
Scalable: From 6-well up to 96-well plates

Works with Stem Cell. Primary Cells, and Cell Lines



Osteoblast cell lines, Clonetics™ NHOst — Normal Human Osteoblasts and osteoblast differentiated Poietics™ hMSC Human Mesenchymal Stem Cells were evaluated for mineralization with the Osteolmage™ Mineralization Assay on a 96-well plate reader.

Detects Mineralization With Time



NHOst — Normal Human Osteoblasts were seeded at 3,200 cells/well in a 96-well plate. Cells were cultured as undifferentiated control cells or with differentiation factors. Mineralization was quantitated on a plate reader after staining with the Osteolmage* Assay on days 7, 14 and 21

Cat. No.	Description	Size
PA-1503	Osteolmage™ Mineralization Assay	5×96-wells

www.lonza.com/research www.lonza.com/bioassays

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