Jeff Johnson, Ph.D.

Introduction to

Assay Kits

June 2010 marks the 30th anniversary of Cayman Chemical. The initial product offering consisted primarily of prostaglandins, thromboxanes, and leukotrienes. With the subsequent development of specific antibodies to these eicosanoids, EIAs became part of the Cayman product line in 1987. These EIAs centered on quantifying the primary eicosanoids generated from arachidonic acid and included popular assays for PGE_2, TXB_2, 6-keto $PGF_{1\alpha}$, LTB_4 and several others. These assays continue to be at the core of Cayman's EIA and assay kit product line even today.

Over the years, the assay kit product line has grown substantially into what you see encompassed in this catalog. EIAs, although still a vital and central presence, are no longer the only format represented. The introduction of assay kits in the early to mid 1990s for glutathione, nitrate/nitrite, phospholipase(s), and COX-inhibitor screening began the transformation of the kit line into many unique formats. Our offering now boasts approximately 18 enzyme activity assays, 36 cell-based assays, 16 transcription factor assays, and 25 inhibitor screening assays, just to name a few. Technological advances have allowed us to offer assays using readouts such as Fluorescence Polarization, Luminex®, and FRET. In total there are nearly 250 kits represented in these pages.

Since Cayman's incorporation of the first EIAs, accuracy and value continue to be motivating factors behind the development of our assays. Many of the assay kits displayed in this catalog, however, originate from beyond the walls of Cayman and so we want to thank our primary business partners - SPI-BIO (France), Biosense (Norway), Columbia Biosciences (USA), and Originus, Inc. (Ann Arbor) - for contributing to our success and growth. In addition, we thank you, our customer, for using our products as we partner with you to help make research possible.

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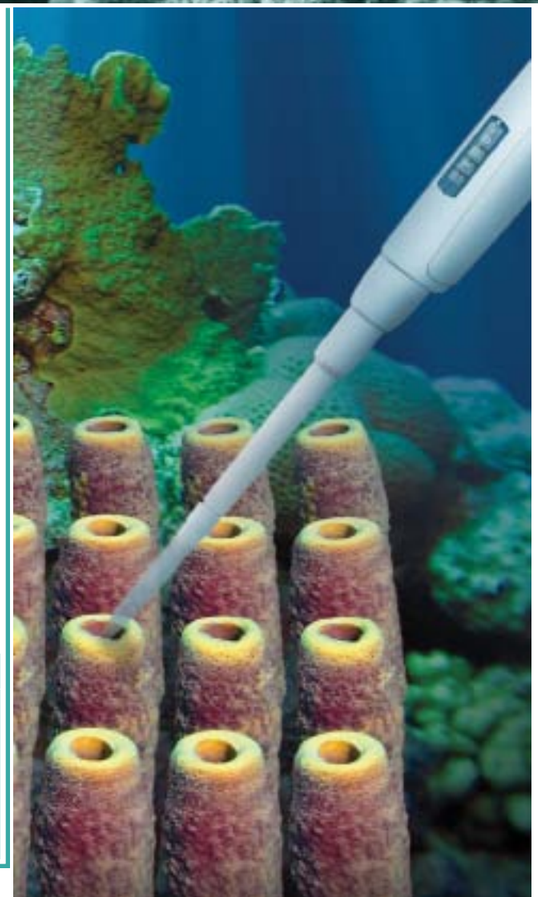
Assay Kits



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Fax: (734) 971-3640
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abbreviations

2-AG	2-Arachidonoyl Glycerol
7-AAD	7-Amino Actinomycin D
AChE	Acetylcholinesterase
AdoHcy	S-Adenosyl Homocysteine
AEA	Arachidonoyl Ethanolamide
COX	Cyclooxygenase
CGRP	Calcitonin Gene-Related Peptide
CRE	cAMP Response Element
CysLT	Cysteinyl Leukotriene
DNA	Deoxyribonucleic Acid
EIA	Enzyme Immunoassay
ELISA	Enzyme-Linked Immunosorbent Assay
FAAH	Fatty Acid Amide Hydrolase
FABP	Fatty Acid Binding Protein
FC	Flow Cytometry
FFA	Free Fatty Acid
FP	Fluorescence Polarization
GPCR	G Protein-Coupled Receptor
HDL	High-Density Lipoprotein
H_2O_2	Hydrogen Peroxide
HTS	High Throughput Screening
IL	Interleukin
JmjC	Jumonji C
LDH	Lactate Dehydrogenase
LDL	Low-Density Lipoprotein
LT	Leukotriene
LO	Lipoxygenase
MDC	Monodansyl Cadaverine
MS	Mass Spectrometry
NADPH	Nicotinamide Adenine Dinucleotide Phosphate
NOS	Nitric Oxide Synthase
NSAID	Non-Steroidal Anti-Inflammatory Drug
PAF	Platelet-Activating Factor
PAF-AH	Platelet-Activating Factor-Acetylhydrolase
PBS	Phosphate Buffered Saline
PE	Phycoerythrin
PG	Prostaglandin
PGDH	Prostaglandin Dehydrogenase
cPGES	Prostaglandin E Synthase (cytosolic)
mPGES	Prostaglandin E Synthase (microsomal)
PK	Protein Kinase
PL	Phospholipase
$cPLA_2$	Calcium Dependent Cytosolic Phospholipase A_2
$iPLA_2$	Calcium-Independent Phospholipase A_2
$sPLA_2$	Secretory Phospholipase A_2
PMNL	Polymorphonuclear Leukocyte
PPAR	Peroxisome Proliferator-Activated Receptor
PUFA	Polyunsaturated Fatty Acid
SAH	S-Adenosyl Homocysteine
SAM	S-Adenosyl Methionine
sEH	Soluble Epoxide Hydrolase
TP	Thromboxane Receptor
TX	Thromboxane
WB	Western Blot

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Enzyme Immunoassays: The Details

Enzyme immunoassays (EIAs), also known as enzyme-linked immunosorbent assays (ELISAs), combine antibody binding with enzymatic detection to quantify molecules of interest. EIAs are easy to perform, require little specialized equipment, and both the experienced lab technician and the research lab novice can learn EIA skills quickly. However, a better understanding of the elements and design of EIAs is useful. This article delves into some of the specific details, as well as describes some properties unique to Cayman's EIAs.

A Better Detection Enzyme Makes a Better Assay

Many commercial EIAs use either horseradish peroxidase (HRP) or alkaline phosphatase (AP) as the enzyme that drives color generation. Cayman's ACE™ EIAs use AChE, an enzyme derived from the electric organ of the electric eel, *Electrophorus electricus*. Acetylcholinesterase has an extremely high turnover number (64,000 per second), which allows for rapid color development in an immunoassay. Unlike HRP, AChE does not suicide inactivate, allowing assays to be redeveloped if they are accidently splashed or spilled. AChE is highly stable under assay conditions, is active over a wide pH range, and tolerates both phosphate and azide.

Competitive EIAs

Competitive EIAs are most commonly used to measure small molecules including lipids, hormones, and small peptides, although larger molecules can also be measured using this format if they are present in high enough concentrations. This type of assay is based on the competition between the analyte of interest and an enzyme-conjugated version of the same analyte (referred to as the tracer) for a limited number of specific antibody binding sites (Figure 1). The concentration of tracer is held constant in all wells while the concentration of analyte varies from well-to-well. As a result, the amount of tracer that can bind to the antibody will be inversely proportional to the amount of analyte in the well – the presence of more analyte means less tracer will be able to bind to the specific antibody. The antibody-analyte (either free or tracer) complex is immobilized by binding to an anti-species IgG antibody coated to the wells of the plate. After sufficient equilibration time, unbound reagents are simply washed away. The developing solution which contains the substrate for AChE and Ellman's reagent is then added to the wells. The reaction product has a distinct yellow color which absorbs strongly at 412 nm.

When performing a competitive EIA, there are several types of wells that must be run on each plate in addition to the wells containing the samples. These types of wells, what they contain, and the information they provide is summarized in the table below.

While it is possible to plot standard curves for a competitive EIA as the concentration of standard (x-axis) versus absorbance (y-axis), it is common practice to instead plot the y-axis value as percent of maximal binding (%B/B_0) (Figure 2). This value is obtained by dividing the absorbance for each well by the absorbance of the maximum binding well (B_0) and multiplying by 100. This method offers the ability to easily compare results between assays performed on two different plates or two different days. While the absolute absorbance may differ from plate to plate or day to day, the %B/B_0 values should be reasonably consistent from one plate to the next.

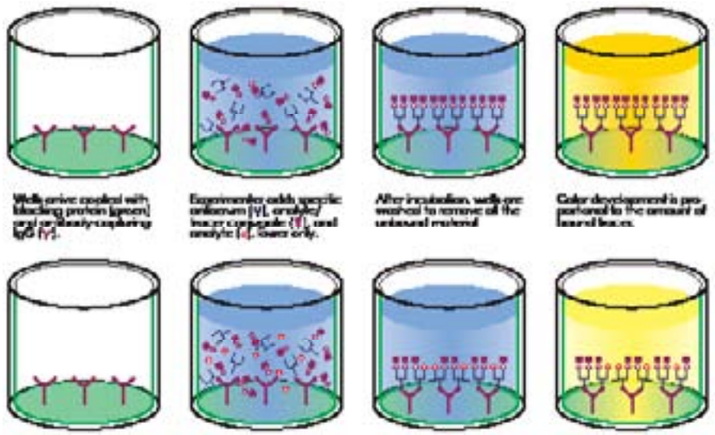


Figure 1. Illustration of a competitive EIA without (above) or with competition (below)

Well Type/Name	Contains	Indicates
Non-Specific Binding (NSB)	Tracer and buffer	How much tracer binds to the wells of the plate in the absence of any specific antibody
Maximum Binding (B_0)	Tracer, antibody, and buffer	Maximum signal obtainable by the antibody and tracer pair in the absence of any analyte
Standard Curve	Tracer, antibody, and a series of known amounts of analyte	An accurate reference across a broad analyte concentration range for determining analyte levels in the sample
Total Activity (TA)	Small amount of tracer (added at the development stage)	Enzyme is active

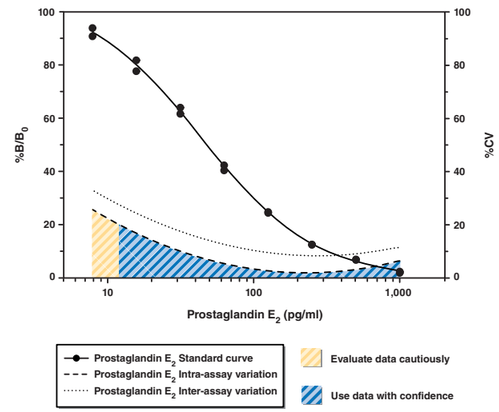


Figure 2. Typical standard curve for Cayman's PGE_2 EIA Kit (Catalog No. 514010)

There are two curve fits that are routinely used for analysis of competitive assays, the four-parameter logistic fit and the log-logit fit. Both curve fits express the standard concentrations in logarithmic fashion on the x-axis. The four-parameter logistic fit uses %B/B_0 on the *y*-axis; analysis of standard curve data using this curve fit results in a sigmoidal-shaped standard curve. The log-logit fit uses logit B/B_0 on the *y*-axis and can be fit with a straight line. The log-logit fit is easy to perform and data can be analyzed in any spreadsheet program. Data analysis tools using the log-logit fit are available on Cayman's website. While the log-logit fit is very convenient, the line fit is sometimes hard to interpret and it is difficult to know where the highest variation in the assay occurs. The sigmoidal curve fit, on the other hand, gives a clear graphical representation of the data that is easily interpretable in terms of where samples can be most accurately analyzed (the linear center portion of the standard curve which is typically between 20-80% B/B_0) and when sample concentrations should be evaluated with more caution (at the ends of the standard curve where it becomes non-linear). In order to provide as much information as possible about the performance of our assays, Cayman determines both the intra-assay and inter-assay Coefficient of Variation (%CV) at all concentrations of the standard curve. For easy reference, the region along the standard curve where the data can be evaluated with confidence is illustrated in blue. %CVs are typically lowest in the center of the curve and increase at the ends of the curve.

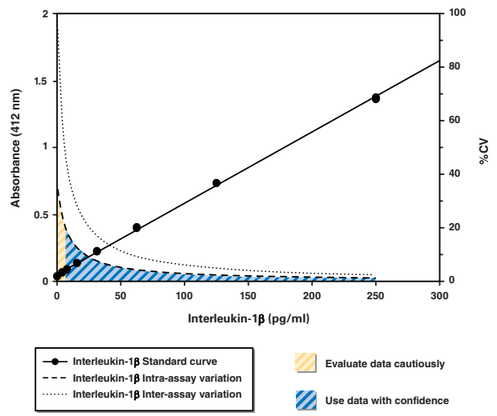


Figure 3. Typical standard curve for Cayman's IL-1β (human) EIA Kit (Catalog No. 583311)

The limit of detection (or sensitivity) of competitive immunoassays can be defined in different ways. While some companies define the sensitivity of their assays using a mathematical formula to estimate a concentration of standard that would give an absorbance greater than that of the NSB, Cayman takes a more conservative approach. We believe that it is important to define sensitivity based on the concentration of analyte where a customer would be truly able to quantify this concentration using the standard curve. Since the majority of competitive immunoassays become non-linear around 80% B/B_0, Cayman defines sensitivity as the concentration of analyte which would result in an absorbance of 80% B/B_0. While this approach often makes Cayman's assays appear on paper to be less sensitive than those of the competition, we feel that it gives customers a more realistic estimation of quantities of analyte that they will be able to accurately measure with our kits.

Immunometric/Sandwich Assays

The second type of solid phase immunoassay, and the most familiar to most researchers, is the immunometric or "sandwich" assay. As the name implies, this assay involves two antibodies which "sandwich" the analyte between them. Because the analyte must be relatively large to allow simultaneous binding of two antibodies, this type of assay is suitable only for proteins and peptides greater than 20 amino acids in length. In most immunometric assays, plates are coated with a capture antibody that is specific to the analyte. This antibody will bind to the analyte present in a sample or standard. A second antibody, called the detection antibody, which recognizes a different epitope on the analyte is also added to the well, resulting in the analyte being "sandwiched" between the two antibodies. This reaction is allowed to come to equilibrium and excess unbound reagents are then washed away. The presence of the detection antibody, which is directly proportional to the amount of analyte present, can be quantified by a few different methods. If the detection antibody was raised in a different species than the coating antibody, it is possible to use an anti-IgG antibody which was raised against IgG of the species that the detection antibody was raised in. If the coating antibody and detection antibody were raised in the same species, then the detection antibody must be tagged in some way. The two most common methods for tagging are biotinylation or direct conjugation of an enzyme to the detection antibody. The biotinylated detection antibody can be measured using a variety of streptavidin-based techniques. Cayman uses both of these approaches for our immunometric assays.

Standard curves for immunometric assays are plotted as the concentration of standard (x-axis) *versus* absorbance (y-axis) (Figure 3). Both axes are usually plotted on a linear scale. Data can sometimes be fit with a straight line, but are generally better fit using a quadratic equation. Sensitivity of Cayman's immunometric assays is defined as the point where the signal obtained with analyte is two times the signal of the non-specific binding.

Additional Considerations

Regardless of the type of immunoassay, there are common aspects in designing and performing an EIA that will ensure the user gets the most from the assay. First, all immunoassays should contain blank wells. Blank wells receive only developing solution and measure the very low absorbance of that solution. Most plate readers will automatically subtract the blank values from the absorbance values obtained for all other wells on the plate. Second, an appropriate set of standards (as well as NSB wells and B_0 wells if appropriate) should be run on each plate. One curve is not sufficient for use with samples on multiple assay plates, even if the plates are run at the same time. Third, all samples should be run at least in duplicate and preferably in triplicate. Additionally, samples should be run at a minimum of two dilutions that fall on the standard curve. The two dilutions should show good correlation in the final calculated concentration of analyte. If they do not show good correlation, it is an indication that something in the samples is interfering with the assay and purification of samples is recommended. Finally, accurate pipetting of small volumes is crucial to obtaining useful data. Cayman sells a practice kit (Catalog No. 10009658) that can used to become familiar with performing a typical competitive EIA before analyzing valuable samples.

Bioactive Peptides

Angiotensin II EIA Kit+ 589301

Please see the **Hypertension** section for full listing on page 23

Arginine Vasopressin EIA Kit 583951

Please see the **Hypertension** section for full listing on page 24

Atriopeptin (rat) EIA Kit+ 589401

Please see the **Hypertension** section for full listing on page 24

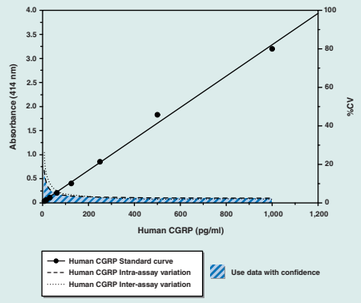
CGRP

CGRP is a 37 amino acid peptide synthesized in the central and peripheral nervous system from a calcitonin/CGRP gene complex. Two isoforms, CGRP-α and CGRP-β, have been described which differ by three amino acids and display similar biological activities. In the CNS, CGRP acts as a neurotransmitter that is released from a subset of small sensory neurons that transmit pain information. In the circulation, CGRP is one of the most potent vasodilators known and may function as a regulator of blood flow.

CGRP (human) EIA Kit+ 589101

Stability: ≥6 months at -20°C **Limit of detection:** <5 pg/ml

1 ea

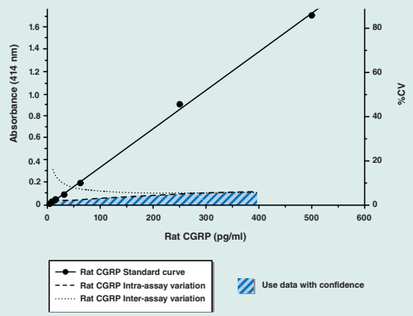


•Also Available: **CGRP Affinity Sorbent+** (489009)

CGRP (rat) EIA Kit+ 589001

Stability: ≥6 months at -20°C **Limit of detection:** <1 pg/ml

1 ea



•Also Available: **CGRP Affinity Sorbent+** (489009)

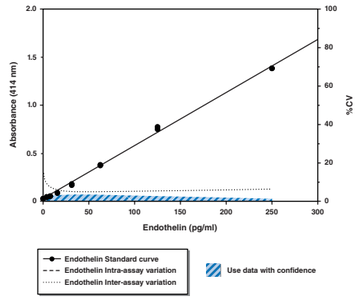
Endothelin EIA Kit 583151

ET

Stability: ≥6 months at -20°C **Limit of Detection:** 7.8 pg/ml

Summary: The endothelin peptide family consists of three isoforms, ET-1 (corresponding to the initially isolated and most predominant isoform), ET-2, and ET-3. ET-1 is a 21 amino acid peptide and is one of the most potent vasoconstrictors currently known. ET-2 displays similar pharmacology to ET-1, whereas ET-3 is a weak vasoconstrictor but more potent inhibitor of platelet aggregation. Cayman's Endothelin Assay offers sensitive and specific analysis of endothelin in serum, plasma, urine, or cell culture media.

96 wells
480 wells



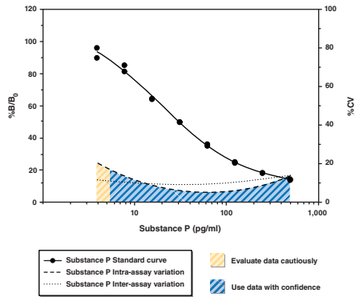
Substance P EIA Kit 583751

Stability: ≥1 year at -20°C

Sensitivity: 50% B/B_0: 32 pg/ml • 80% B/B_0: 8.2 pg/ml

Summary: Substance P is a bioactive 11-amino acid peptide (Arg-Pro-Lys-Pro-Gln-Gln-Phe-Phe-Gly-Leu-Met-amide) that is involved in many physiological processes including pain modulation, smooth muscle contraction, blood pressure control, kidney function, and water homeostasis. Substance P is widely distributed in numerous tissues and body fluids including the central and peripheral nervous syst, gastrointestinal tract, respiratory tract, visual system, and circulatory system.

96 wells
480 wells



•Also Available: **Substance P EIA Kit (Solid Plate)** (583751.1)

SPI-BIO products are available through Cayman Chemical only within North & South America and Asia; elsewhere contact SPI-BIO.

Cell-Based Assays

7-AAD Cell Viability Assay Kit 10009856

7-Amino Actinomycin D

Stability: ≥1 year at 4°C **Detection Method(s):** FC

Summary: 7-AAD is a fluorescent dye which is excluded from live cells but penetrates dead or damaged cells to label DNA. Although 7-AAD fluorescence is less intense than that of propidium iodide, it exhibits a higher wavelength emission maximum (excitation at 488 nm, emission at 650 nm) and thus has minimal spectral overlap with PE or FITC. This makes 7-AAD preferable as a viability marker when FITC and/or PE are used simultaneously to label surface or intracellular antigens.

1 ea

7-AAD/CFSE Cell-Mediated Cytotoxicity Assay Kit 600120

7-Amino Actinomycin D, 5-(6)-Carboxyfluorescein Diacetate Succinimidyl Ester

Stability: ≥1 year at -20°C **Detection Method(s):** FC

Summary: Cayman's 7-AAD/CFSE Cell-Mediated Cytotoxicity Assay employs CFSE to label target cells and 7-AAD to label dead cells. The labeling distinguishes four populations of cells: 1) living target cells in green, 2) dead target cells in green and red, 3) dead effector cells in red, and 4) live effector cells, which remain unstained. Thus, the cytotoxicity of the effector cells can be evaluated quantitatively.

1 ea

Adipogenesis Assay Kit 10006908

Please see the **Endocrinology and Metabolism** section for full listing on page 16

Adipolysis Assay Kit 10009381

Please see the **Endocrinology and Metabolism** section for full listing on page 17

Aldehyde Site (DNA and Protein) Detection Kit 600170

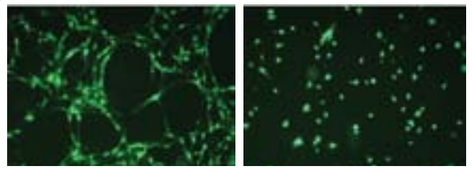
Please see the **Oxidative Injury** section for full listing on page 38

In Vitro Angiogenesis Assay Kit 10009964

Stability: ≥1 year at 4°C **Detection Method(s):** Fluorescence microscope

Summary: Cayman's *In Vitro* Angiogenesis Assay uses a one-step model to study regulators of angiogenesis. Cell survival is improved compared to the other assays by use of a modified extracellular matrix that has been validated in both short-term (2-3 days) and long-term (up to ten days) experiments. Cayman's *In Vitro* Angiogenesis Assay includes PMA and JNJ-10198409 as controls for stimulation and inhibition of angiogenesis, respectively, as well as the fluorescent dye Calcein AM for visualization of cell organization.

1 ea



Inhibition of network formation by JNJ-10198409. *Panel A:* CAPE cells treated with 0.064 µM PMA or *Panel B:* 0.064 µM PMA + 0.3 µM JNJ-10198409. Cells were stained with Calcein AM.

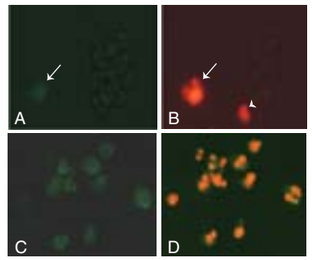
Apoptotic Blebs Assay Kit 10010750

Stability: ≥6 months at 4°C

Detection Method(s): FC, Fluorescence microscope, and Plate reader

Summary: Cayman's Apoptotic Blebs Assay employs a recombinant protein derived from single chain variable fragments (scFv) of an antibody which preferentially binds to the autoantigen in membrane blebs of apoptotic cells. This recombinant protein is fused to protein A allowing visualization using fluorescein-conjugated rabbit IgG. This assay is specific for cells in the execution stage of apoptosis.

96 wells



Panels A and B: Apoptotic blebs in green (A) and dead cells in red (B). *Panels C and D:* Apoptotic blebs in green (C) and dead cells from apoptosis in orange (D) in staurosporine-treated cells.

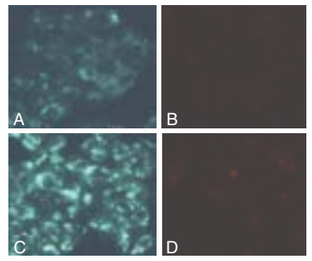
Autophagy/Cytotoxicity Dual Staining Kit 600140

Stability: ≥6 months at -20°C

Detection Method(s): Fluorescence microscope

Summary: Cayman's Autophagy/Cytotoxicity Dual Staining provides a convenient tool for studying the regulation of autophagy and cytotoxicity at the cellular level. The kit employs MDC, an autofluorescent substance that is incorporated into multilamellar bodies by both an ion trapping mechanism and the interaction with membrane lipids, as a probe for detection of autophagic vacuoles in cultured cells. Propidium iodide is used as a marker of cell death. Tamoxifen, a known inducer of autophagy, is included as a positive control.

1 ea

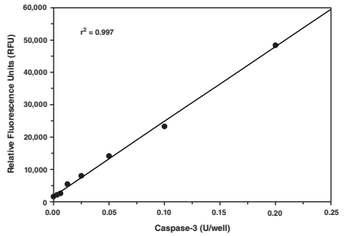


Tamoxifen increases autophagy but not cell death in HepG2 cells as measured by fluorescence microscopy. *Panel A:* MDC staining of HepG2 cells treated with vehicle. *Panel B:* Propidium iodide staining of HepG2 cells treated with vehicle. *Panel C:* MDC staining of HepG2 cells treated with 10 µM Tamoxifen. *Panel D:* Propidium iodide staining of HepG2 cells treated with 10 µM Tamoxifen.

Caspase-3 Fluorescence Assay Kit 10009135

Stability: ≥6 months at -20°C
Detection Method(s): Plate reader
Summary: Caspase-3 plays a central role in the execution of apoptosis. Cayman's Caspase-3 Fluorescence Assay employs a specific caspase-3 substrate, N-Ac-DEVD-N'-MC-R110, which upon cleavage by active caspase-3, generates a highly fluorescent product that can be measured using excitation and emission wavelengths of 485 and 535 nm, respectively. Active caspase-3 is included in the kit as a positive control or as a quantitative standard.

96 wells



Cell Cycle Phase Determination Kit 10009349

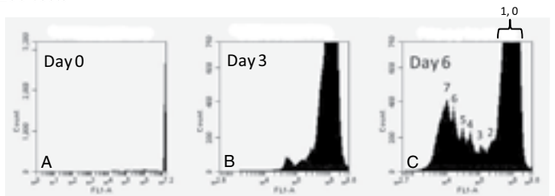
Stability: ≥6 months at -20°C **Detection Method(s):** FC
Summary: Cayman's Cell Cycle Phase Determination Kit provides an easy to use tool for studying the induction and inhibition of cell cycle progression in any cell suspension sample. The assay involves the fixation and permeabilization of the cells of interest, making possible the staining of DNA within intact cells by propidium iodide. This kit will allow the investigator to determine the percentage of cells in a given sample that are within G_1/G_0, G_2, or S phase at the time of fixation, as well as to quantify cells in the sub-G_1 phase prior to apoptosis by FC.

100 tests

CFSE Cell Division Assay Kit 10009853

Stability: ≥1 year at -20°C **Detection Method(s):** FC
Summary: Carboxyfluorescein diacetate succinimidyl ester (CFDA-SE) is a novel cell-tracing fluorescent dye used to examine the proliferative activity of cells by the labeling of a parent generation and the inheritance of the label by daughter generations. CFDA-SE diffuses into cells, where the acetate groups on the molecule are cleaved to yield a highly fluorescent derivative (CFSE) that is retained in the cell and can be detected by FC. Cell division results in sequential halving of fluorescence, and up to eight divisions can be monitored before the fluorescence is decreased to the background fluorescence of unstained cells.

100 tests



Human peripheral blood lymphocyte proliferation. *Panel A:* CFSE-labeled lyphocytes Day 0. *Panel B:* CFSE-labeled lymphocytes Day 3 *Panel C:* CFSE-labeled lymphocytes Day 6.

Cholesterol Cell-Based Detection Assay Kit 10009779

Please see the **Lipids** section for full listing on page 28

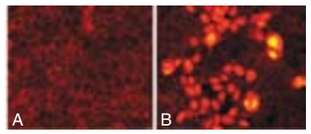
ChREBP Cell-Based Translocation Assay Kit 10010060

Please see the **Transcription Factor** section for full listing on page 56

ERK/MAPK (Phospho-Thr^{202}/Tyr^{204}) Cell-Based Phosphorylation/Translocation Assay Kit 10010549

Stability: ≥1 year at -20°C **Detection Method(s):** Fluorescence microscope
Summary: Cayman's ERK/MAPK (Phospho-Thr^{202}/Tyr^{204}) Cell-Based Phosphorylation/Translocation Assay provides the tools necessary to study ERK/MAPK phosphorylation and translocation within whole cells. The kit contains a phospho-specific ERK/MAPK (Phospho-Thr^{202} and Tyr^{204}) primary antibody together with a Dylight™ (product of Thermo Scientific) conjugated secondary antibody in a ready-to-use format. Tamoxifen, which has been shown by scientists at Cayman Chemical to cause the translocation of phosphorylated ERK/MAPK (Phospho-Thr^{202}/Tyr^{204}) between the cytoplasm and nuclear compartments, is included as a positive control.

1 ea

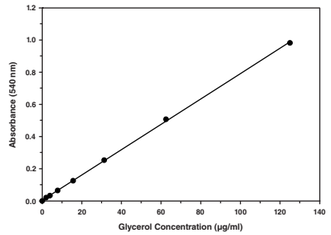


Tamoxifens induce the translocation of ERK/MAPK (Phosopho-Thr^{202}/Tyr^{204}) from the cytoplasm to the nucleus in MCF-7 cells. *Panel A:* Cells treated with vehicle. *Panel B:* Cells treated with 20 µM tamoxifen for 20 minutes.

Glycerol Cell-Based Assay Kit 10011725

Stability: ≥1 year at -20°C **Detection Method(s):** Plate reader
Summary: Cayman's Glycerol Cell-Based Assay provides a convenient tool for studying triglyceride/fatty acid cycling and its regulation in adipocytes or hepatocytes. This kit will allow investigators to screen compounds involved in lipid storage and metabolism. Chloroquine is included in the kit as a positive control for screening pharmaceuticals that induce lipid droplet accumulation and free glycerol release from hepatocytes.

1 ea



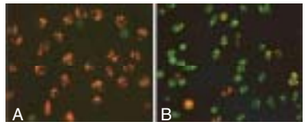
Hydrogen Peroxide Cell-Based Assay Kit 600050

H_2O_2
Please see the **Oxidative Injury** section for full listing on page 40

JC-1 Mitochondrial Membrane Potential Assay Kit 10009172

Stability: ≥6 months at -20°C
Detection Method(s): FC, Fluorescence microscope, and Plate reader
Summary: Mitochondrial membrane potential, Δψm, is an important parameter of mitochondrial function that is used as an indicator of cell health. JC-1 is a lipophilic, cationic dye that can selectively enter into mitochondria and reversibly change color from green to red as the membrane potential increases. In healthy cells with high mitochondrial Δψm, JC-1 spontaneously forms complexes known as J-aggregates with intense red fluorescence. On the other hand, in apoptotic or unhealthy cells with low Δψm, JC-1 remains in the monomeric form, which shows only green fluorescence. Cayman's JC-1 Mitochondrial Membrane Potential Assay provides all the necessary reagents, as well as complete instructions, for analysis of mitochondrial integrity in whole cells.

100 tests

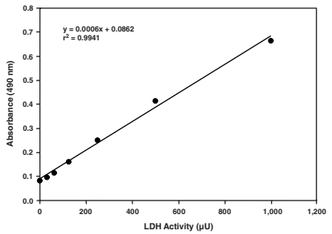


Effect of staurosporine on mitochondrial potential in Jurkat cells. *Panel A:* Untreated cells show most of cells had strong J-aggregation (red). *Panel B:* Staurosporine-treated cells show a majority of cells stained green due to low ΔΨm.

LDH Cytotoxicity Assay Kit 10008882

Lactate Dehydrogenase Cytotoxicity
Stability: ≥1 year at -20°C **Detection Method(s):** Plate reader
Summary: LDH is a soluble cytosolic enzyme that is released into the culture medium following loss of membrane integrity resulting from either apoptosis or necrosis. LDH activity, therefore, can be used as an indicator of cell membrane integrity and serves as a general means to assess cytotoxicity resulting from chemical compounds or environmental toxic factors. Cayman's LDH Cytotoxicity Assay measures LDH activity present in culture medium using a coupled two-step reaction producing a highly-colored formazan dye which absorbs strongly at 490-520 nm.

96 wells
480 wells



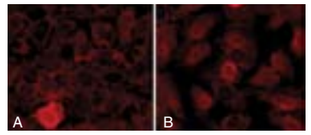
Lipid Droplets Fluorescence Assay Kit 500001

Please see the **Lipids** section for full listing on page 29

p38 MAPK (Phospho-Thr^{180}/Tyr^{182}) Cell-Based Phosphorylation/Translocation Assay Kit 10010374

Stability: ≥1 year at -20°C **Detection Method(s):** Fluoresence microscope
Summary: p38 MAPK is activated by phosphorylation at Thr^{180} and Tyr^{182} in response to both inflammatory cytokines and stress. The subcellular location of p38 following stimulation is not well understood. Cayman's p38 Cell-Based Phosphorylation/Translocation Assay provides a highly specific phospho-p38 MAPK (phospho-Thr^{180} and Tyr^{182}) primary antibody together with a Dylight™ (product of Thermo Scientific) conjugated secondary antibody in a ready-to-use format. Thrombin, for treatment of cells, is included as a positive control.

1 ea



Thrombin-induced translocation of p38 MAPK (Phospho-Thr^{180}/Tyr^{182}) in HeLa cells. HeLa cells were treated with vehicle *Panel A:* or thrombin 10 U/ml *Panel B:* for three hours, then fixed and stained with p53 MAPK (Phospho-Thr^{180}/Tyr^{182}) primary antibody and a goat anti-rabbit antibody conjugated to DyLight™ 549.

p53 Cell-Based Activation/Translocation Assay Kit 600008

Please see the **Transcription Factor Assays** section for full listing on page 58

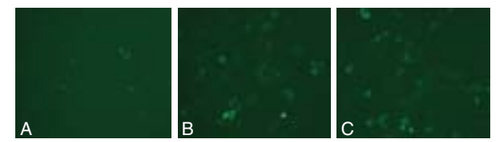
p53 Total and p53 (Phospho-Ser^{392}) Dual Staining Assay Kit 600060

Please see the **Transcription Factor Assays** section for full listing on page 58

Phagocytosis Assay Kit (IgG FITC) 500290

Stability: ≥6 months at 4°C
Detection Method(s): FC and Fuoresence microscope
Summary: Cayman's Phagocytosis Assay (IgG FITC) employs latex beads coated with fluorescently-labeled rabbit-IgG as a probe for the identification of factors regulating the phagocytic process *in vitro*. The engulfed fluorescent-beads can be detected by fluoresence microscopy or flow cytometry.

1 ea

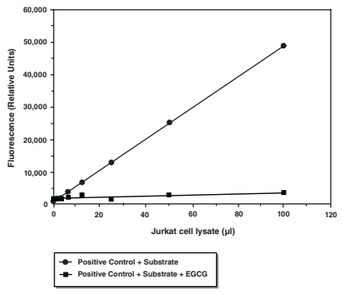


Differentiation of THP-1 cells into phagocytic cells. THP-1 cells treated with either vehicle *Panel A:*, 0.16 µM PMA *Panel B:*, or 1.6 µM PMA *Panel C*: and at the same time loaded with Latex Beads-Rabbit IgG-FITC Complex.

20S Proteasome Assay Kit 10008041

Stability: ≥6 months at -80°C **Detection Method(s)** Plate reader
Summary: The proteasome is a multicatalytic proteinase complex that is involved in the selective degradation of intracellular proteins. Proteasome inhibitors exhibit anti-inflammatory and antiproliferative effects. Cayman's 20S Proteasome Assay employs a specific 20S substrate, SUC-LLVY-AMC which, upon cleavage by the active enzyme, generates a highly fluorescent product with emission at 480 nm.

1 ea



Soluble Epoxide Hydrolase Cell-Based Assay Kit 600090

Please see the **Hypertension** section for full listing on page 24

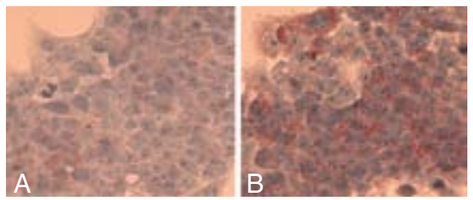
SREBP-2 Cell-Based Translocation Assay Kit 10009239

Please see the **Transcription Factor** section for full listing on page 60

Steatosis Colorimetric Assay Kit 10012643

Stability: ≥1 year at 4°C **Detection Method(s):** Plate reader and Light microscope
Summary: Steatosis, also known as fatty liver, is a pathological process characterized by abnormal accumulation of lipid within cells. Cayman Chemical's Steatosis Colorimetric Assay provides a convenient tool for evaluating the steatosis risk of drug candidates. In this assay, Oil Red O is used to stain neutral lipids in hepatocytes. Lipid accumulation is then quantified using a plate reader after the dye is extracted from the lipid droplets. Chloroquine is included in the kit as a positive control.

1 ea

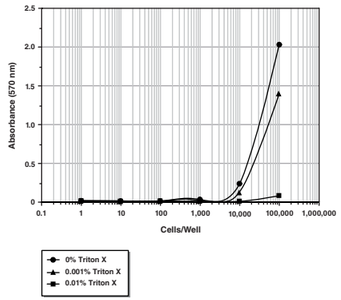


Effect of chloroquine on lipid droplet accumulation in HepG2 cells. *Left Panel:* HepG2 cells treated with vehicle. *Right Panel:* HepG2 cells treated with 25 µM chloroquine.

MTT Cell Proliferation Assay Kit 10009365

Stability: ≥6 months at 4°C **Detection Method(s):** Plate reader
Summary: Cayman's MTT Proliferation Assay provides an easy to use tool for studying the induction and inhibition of cell proliferation in any *in vitro* model. This kit will also allow investigators to screen drug candidates involved in cell cycle regulation. In this assay, MTT is taken up by cells through the plasma membrane and then reduced to formazan by intracellular NAD(P)H-oxidoreductases. The assay can be performed in 3-4 hours with no wash steps in a 96-well plate format.

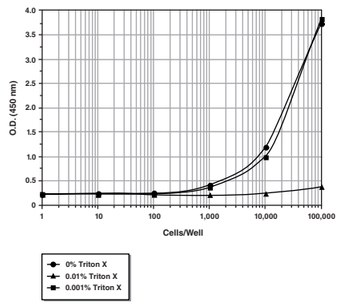
480 wells
2,400 wells



WST-1 Cell Proliferation Assay Kit 10008883

Stability: ≥1 year at -20°C **Detection Method(s):** Plate reader
Summary: Cayman's WST-1 Proliferation Assay is based on the reduction of tetrazolium salt WST-1 to soluble formazan by electron transport across the plasma membrane of dividing cells. This kit will also allow investigators to screen drug candidates involved in cell cycle regulation.

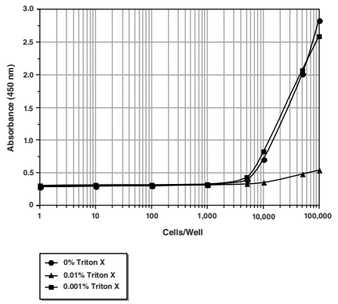
96 wells
480 wells
4,800 wells



WST-8 Cell Proliferation Assay Kit 10010199

Stability: ≥1 year at -20°C **Detection Method(s):** Plate reader
Summary: Cayman's WST-8 Cell Proliferation Assay provides an easy to use tool for studying the induction or inhibition of cell proliferation in any *in vitro* model. The assay is based on the extracellular reduction of WST-8 by NADH produced in the mitochondria *via* trans-plasma membrane electron transport and an electron mediator. Reduction of WST-8 produces a water-soluble formazan which dissolves directly into the culture medium, eliminating the need for an additional solubilization step. WST-8 is more stable and less cytotoxic than the other tetrazolium salts, making it especially useful for longer incubation periods. The detection sensitivity is higher than that for other tetrazolium salts.

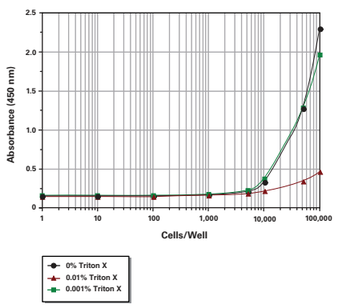
96 wells
480 wells
4,800 wells



XTT Cell Proliferation Assay Kit 10010200

Stability: ≥ 1 year at -20°C **Detection Method(s):** Plate reader
Summary: Cayman's XTT Cell Proliferation Assay provides an easy to use tool for studying induction and inhibition of cell proliferation in any *in vitro* model. The assay is based on the extracellular reduction of XTT by NADH produced in the mitochondria *via trans*-plasma membrane electron transport and an electron mediator. Reduction of XTT produces a water-soluble formazan dye which dissolves directly into the culture medium, eliminating the need for an additional solubilization step. This kit will allow investigators to screen drug candidates involved in cell cycle regulation.

96 wells
480 wells
4,800 wells



Chemical Libraries

Cayman Chemical offers chemical libraries for drug discovery and development. The libraries are produced from our inventory of high-quality and high-purity compounds and presented in a 96-well format. Screening hits may be purchased individually for activity confirmation and further analysis. Many items are also available in bulk or on a custom basis at a reasonable cost. In addition, Cayman produces custom libraries using a customer-designated format and specifications. Structural information for our diverse collection of compounds is available for download and may be used for virtual screening and hit-seeking free of charge.

Fatty Acid Screening Library (96-Well) 10504

Stability: ≥6 months at -20°C
Summary: This screening plate contains a variety of fatty acids with diverse biological activities as a 10 mM solution in DMSO.

50 µl
100 µl
200 µl

Prostaglandin Screening Library I (96-Well) 10501

Stability: ≥6 months at -20°C
Summary: This screening plate contains a wide range of PGs in the "F-series" configuration (9,11-hydroxy PGs) as a 2 mM solution in DMSO.

50 µl
100 µl
200 µl

Prostaglandin Screening Library II (96-Well) 10502

Stability: ≥6 months at -20°C
Summary: This screening plate contains wide range PGs in the "D and E-series" configuration (9-keto, 11-keto, 9-hydroxy, and 11-hydroxy PGs) as a 2 mM solution in DMSO.

50 µl
100 µl
200 µl

Prostaglandin Screening Library III (96-Well) 10503

Stability: ≥6 months at -20°C
Summary: This screening plate contains wide range PGs in the "A and J-series" as a 2 mM solution in DMSO.

50 µl
100 µl
200 µl

Bio-active Lipid 1 Screening Library (96-Well) 10506

This screening library consists of 11 plates and contains 928 compounds that include prostaglandins, thromboxanes , cannabinoids, D-*myo*-inositol-phosphates, phosphatidylinositol phosphates, sphingolipids, inhibitors, receptor antagonists, ceramide derivatives, and several other complex polyunsaturated fatty acids. This collection is ideal for prostanoid or other G protein-coupled receptor screening, target validation, secondary screening, validating new assays, and for routine pharmacological applications. The library is provided in a 96-well format as 1.0 mM solution in DMSO.

100 µl
500 µl

Bio-active Lipid 2 Screening Library (96-Well) 10507

This screening library consists of 3 plates and contains 240 compounds that include prostaglandins, isoprostanes, thromboxanes, leukotrienes, lipoxins, and several other complex polyunsaturated fatty acids. This collection is deal for prostanoid or other G protein-coupled receptor screening, target validation, secondary screening, validating new assays, and for routine pharmacological applications. The library is provided in a 96-well format as 0.1 mM solution in DMSO.

100 µl

Chromatin Modification

Demethylase (Jumonji-type) Activity Assay Kit 700390

Stability: ≥6 months at -80°C
Summary: Lysine demethylases containing JmjC domains produce formaldehyde following 2-oxoglutarate-dependent demethylation. Cayman's Demethylase (jumonji-type) Activity Assay provides a fluorescence-based method for assaying JmjC-mediated demethylase activity from cell lysates or purified enzyme preparations. The assay is based on the production of formaldehyde during the demethylation of a methylated peptide substrate. Cyclization of formaldehyde and acetoacetanilide in the presence of ammonia gives a fluorescent product for quantitation.

96 wells

Demethylase (LSD1-type) Activity Assay Kit 700400

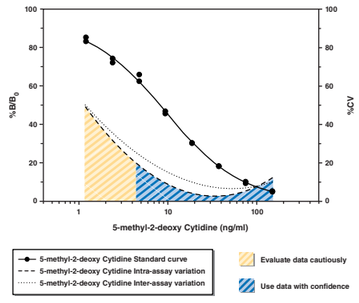
Stability: ≥6 months at -80°C
Summary: Lysine demethylases containing amine oxidase domains, including the lysine-specific demethylases LSD1 and LSD2, specifically demethylate histone H3 at lysine 4 in a FAD-dependent reaction. Cayman's Demethylase (LSD1-type) Activity Assay provides a fluorescence-based method for assaying LSD-type demethylase activity from cell lysates or purified enzyme preparations. The assay is based on the production of formaldehyde during demethylation of lysine 4 on a peptide corresponding to the first 21 amino acids of the N-terminal tail of histone H3. Cyclization of formaldehyde and acetoacetanilide in the presence of ammonia gives a fluorescent product for quantitation.

96 wells

DNA Methylation ElA Kit 589324

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: ~12 ng/ml • 80% B/B_0: ~3 ng/ml
Summary: DNA methylation is an important epigenetic process regulating gene expression. Methylation occurs on carbon 5 of 2-deoxy cytidine yielding the modified base 5-methyl-2-deoxy cytidine. Methylation results in long-term silencing of genes, while unmethylated regions of DNA can be actively transcribed. Cayman's DNA Methylation EIA is a competitive assay that can be used for quantification of 5-methyl-2-deoxy cytidine in urine, culture supernatants, plasma, and other sample matrices.

96 wells
480 wells



•Also Available: **DNA Methylation EIA Kit (Solid Plate)** (589325)

HAT Inhibitor Screening Assay Kit 10006515

Histone Acetyltransferase
Stability: ≥6 months at -20°C
Summary: Cayman's HAT Inhibitor Screening Assay provides a fast, fluorescence-based method for evaluating pCAF HAT inhibitors. The procedure requires only three easy steps, all performed in the same microplate resulting in formation of a highly fluorescent product that is detected using excitation and emission wavelengths of 360-390 and 450-470 nm, respectively.

96 wells

HDAC Activity Assay Kit 10011563

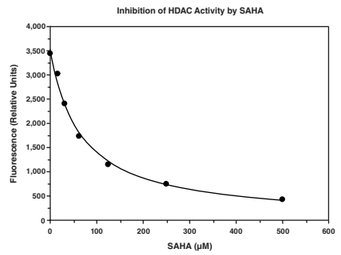
Stability: ≥6 months at -80°C
Summary: Cayman's HDAC Activity Assay provides a fast, fluorescence-based method for measuring Class I and II HDAC activity. The procedure requires only two easy steps, both performed in the same microplate. The fluorescent reaction product is analyzed using a plate reader with excitation wavelengths between 340-360 nm and emission wavelengths between 440-465 nm.

96 wells

HDAC Cell-Based Activity Assay Kit 600150

Stability: ≥6 months at -80°C
Summary: Cayman's HDAC Cell-Based Assay provides a tool for studying HDAC activity modulators in whole cells. By using a cell-permeable HDAC substrate, the activity of various protein lysine-specific deacetylases, including HDAC1-containing complexes, can be measured in intact cells in a simple and homogenous manner. This assay compliments Cayman's HDAC Activity Assay (Catalog No. 10011563), which uses a nuclear extract rather than whole cells for the assay.

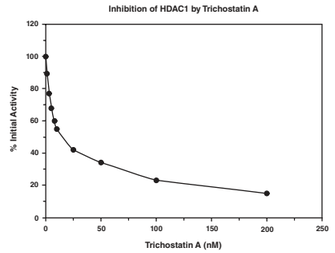
96 wells



HDAC1 Inhibitor Screening Assay Kit 10011564

Stability: ≥6 months at -80°C
Summary: Cayman's HDAC1 Inhibitor Screening Assay provides a fast, fluorescence based method for screening HDAC1 inhibitors. The procedure requires only two easy steps, both performed in the same microplate. The fluorescent reaction product is analyzed using excitation wavelengths between 340-360 nm and emission wavelengths between 440-465 nm. Sufficient purified HDAC1 is provided for 100 tests.

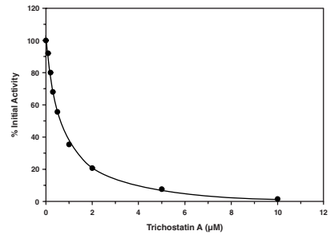
96 wells



HDAC8 Inhibitor Screening Assay Kit 700230

Stability: ≥6 months at -80°C
Summary: Human HDAC8 is a class I HDAC and has been identified in a variety of human cancer tissues. Cayman's HDAC8 Inhibitor Screening Assay provides a fluorescence-based method for screening HDAC8 inhibitors. The procedure requires only two easy steps, both performed in the same microplate. The fluorescent reaction product is analyzed with an excitation wavelength between 350-360 nm and an emission wavelength between 450-465 nm. Sufficient HDAC8 is provided for 100 tests.

96 wells



JMJD2A Inhibitor Screening Assay Kit 700360

Jumonji Domain Containing 2A, Lysine-specific Demethylase 4A, KDM4A, KIAA0677
Stability: ≥6 months at -20°C
Summary: JMJD2A is a JmjC histone demethylase that catalyzes the demethylation of di- and tri-methylated lysine 9 and lysine 36 of histone H3. Cayman's JMJD2A Inhibitor Screening Assay Kit is based on the multistep reaction in which JMJD2A first produces formaldehyde during the demethylation of the trimethylated peptide substrate, histone H3 trimethyl lys9. Cyclization of formaldehyde and acetoacetanilide in the presence of ammonia gives a fluorescent product for quantitation.

96 wells

JMJD2D Inhibitor Screening Assay Kit 700370

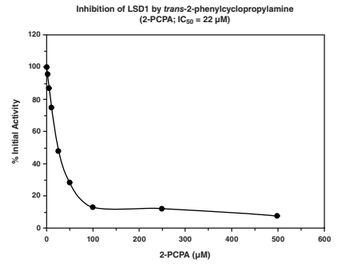
Jumonji Domain Containing 2D, KDM4D, Lysine=specific Demethylase 2D
Stability: ≥6 months at -20°C
Summary: JMJD2D is a JmjC histone demethylase that catalyzes the demethylation of mono-, di-, and tri-methylated lysine 9 of histone H3. Cayman's JMJD2D Inhibitor Screening Assay Kit is based on the multistep reaction in which JMJD2D first produces formaldehyde during the demethylation of the trimethylated peptide substrate, histone H3 trimethyl lys9. Cyclization of formaldehyde and acetoacetanilide in the presence of ammonia gives a fluorescent product for quantitation.

96 wells

LSD1 Inhibitor Screening Assay Kit 700120

Lysine-Specific Demethylase 1
Stability: ≥6 months at -80°C
Summary: LSD1 is a histone demethylase whose actions on specific lysine residues alter transcription of chromosomal DNA. It also inhibits the tumor suppressor activity of p53 by demethylating a specific lysine residue. Cayman's LSD1 Inhibitor Screening Assay is based on the multistep enzymatic reaction in which LSD1 first produces H_2O_2 during the demethylation of lysine 4 of a histone 3 peptide. In the presence of horseradish peroxidase, H_2O_2 reacts with ADHP to produce the highly fluorescent compound resorufin.

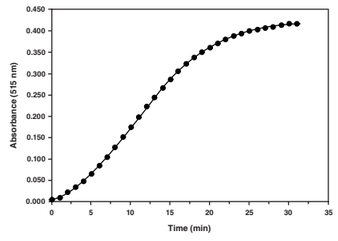
96 wells



Methyltransferase Colorimetric Assay Kit 700140

Stability: ≥6 months at -80°C
Summary: Cayman's MT Colorimetric Assay is a continuous enzyme-coupled assay that can continuously monitor SAM-dependent MT activities. The removal of the methyl group from SAM generates AdoHcy, which is rapidly converted to urate and H_2O_2 by an enzyme mixture provided in the kit. H_2O_2 is measured with the colorimetric reagent 3,5-dichloro-2-hydroxybenzenesulfonic acid. The assay can be used with any purified SAM-dependent MT.

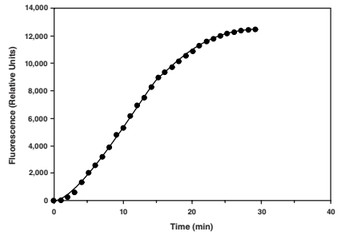
96 wells



Methyltransferase Fluorometric Assay Kit 700150

Stability: ≥6 months at -80°C
Summary: Cayman's MT Fluorometric Assay is a continuous enzyme-coupled assay that can continuously monitor SAM-dependent MTs. The removal of the methyl group from SAM generates AdoHcy (SAH), which is rapidly converted to urate and H_2O_2 by an enzyme mixture provided in the kit. The reaction between H_2O_2 and ADHP produces the highly fluorescent compound resorufin. The assay can be used with any purified SAM-dependent MT.

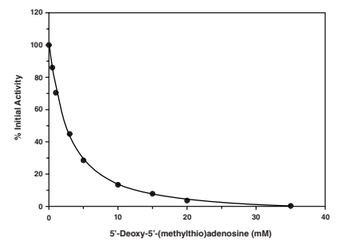
96 wells



SET7/9 Methyltransferase Inhibitor Screening Assay Kit 700270

Stability: ≥6 months at -80°C
Summary: SET7/9 is a MT that acts on various substrates including histone 3 at lysine residue 4, p53, and the transcription factor TAF 10. In Cayman's SET7/9 MT Inhibitor Screening Assay the transfer of the methyl group from SAM by SET7/9 to the acceptor peptide (TAF 10) generates AdoHcy (SAH), which is rapidly converted to urate and H_2O_2 using an enzyme mixture provided in the kit. A subsequent reaction between H_2O_2 and ADHP produces the highly fluorescent compound resorufin.

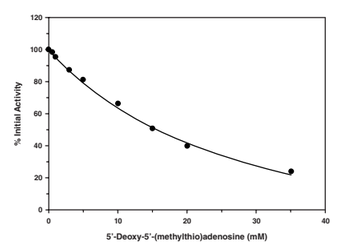
96 wells



SET8 Methyltransferase Inhibitor Screening Assay Kit 700350

PR-Set7, SETD8, SET domain-containing (lysine methyltransferase) 8
Stability: ≥6 months at -80°C
Summary: SET Domain-containing Protein 8 is a MT that selectively mono-methylates histone H4 at lysine residue 20 (H4K20). In Cayman's SET8 MT Inhibitor Screening Assay, the transfer of the methyl group from SAM by SET8 to the acceptor peptide H4K20 generates AdoHcy (SAH), which is rapidly converted to urate and H_2O_2 using an enzyme mixture provided in the kit. A subsequent reaction between H_2O_2 and ADHP produces the highly fluorescent compound resorufin.

96 wells



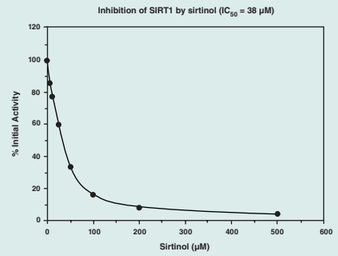
SIRT Direct Fluorescent Screening Assay Kits

The sirtuins represent a distinct class of trichostatin A-insensitive lysyl-deacetylases (class III HDACs) that catalyze a reaction coupling lysine deacetylation to the formation of nicotinamide and O-acetyl-ADP-ribose. Cayman's Direct Fluorescent Screening Assay Kits provide a fluorescence-based method for screening SIRT inhibitors or activators. The procedure requires only two easy steps, both performed in the same microplate. In the first step, the substrate is incubated with human recombinant SIRT along with its cosubstrate NAD^+. Deacetylation sensitizes the substrate such that treatment with the developer in the second step releases a fluorescent product.

SIRT1 Direct Fluorescent Screening Assay Kit 10010401

Stability: ≥6 months at -80°C

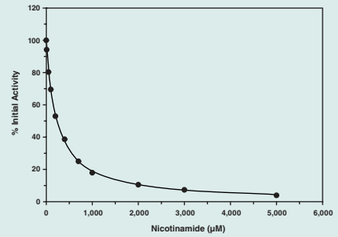
96 wells



SIRT2 Direct Fluorescent Screening Assay Kit 700280

Stability: ≥6 months at -80°C

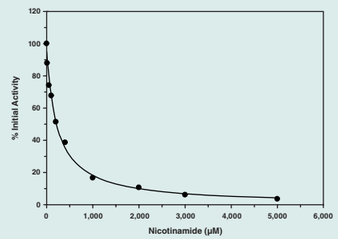
96 wells



SIRT3 Direct Fluorescent Screening Assay Kit 10011566

Stability: ≥6 months at -80°C

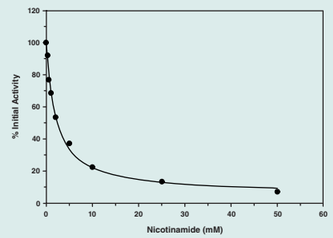
96 wells



SIRT6 Direct Fluorescent Screening Assay Kit 700290

Stability: ≥6 months at -80°C

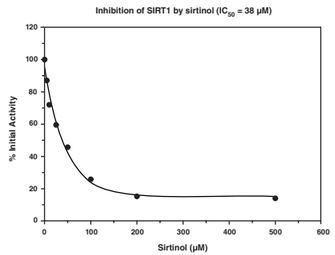
96 wells



SIRT1 FRET-Based Screening Assay Kit 10010991

Stability: ≥6 months at -80°C
Summary: Cayman's SIRT1 FRET-based Screening Assay provides a novel fluorescence-based method for screening SIRT1 inhibitors or activators. The substrate, which is coupled to a fluorophore and quencher, is first incubated with human recombinant SIRT1. Deacetylation sensitizes the substrate such that treatment with a developer separates the quencher and fluorophore resulting in bright fluorescence.

96 wells

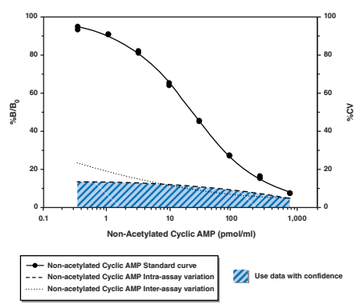


Cyclic Nucleotides

Cyclic AMP EIA Kit 581001

Adenosine 3',5'-cyclic mononucleotide, cAMP
Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 20 pmol/ml (non-acetylated); 0.5 pmol/ml (acetylated)
80% B/B_0: 3 pmol/ml (non-acetylated); 0.1 pmol/ml (acetylated)

96 wells
480 wells

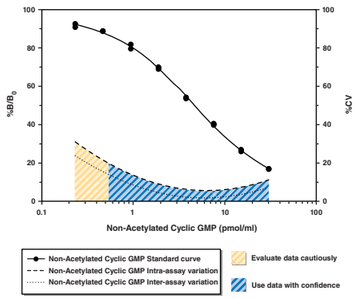


•Also Available: **Cyclic AMP EIA Kit (Solid Plate)** (581001.1)

Cyclic GMP EIA Kit 581021

cGMP, Guanosine 3',5'-cyclic mononucleotide
Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 5.2 pmol/ml (non-acetylated); 0.46 pmol/ml (acetylated)
80% B/B_0: 1 pmol/ml (non-acetylated); 0.1 pmol/ml (acetylated)

96 wells
480 wells



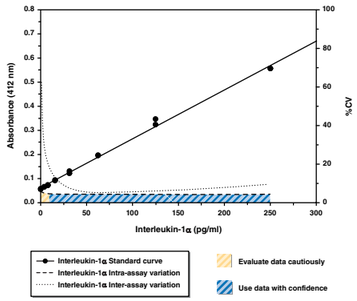
•Also Available: **Cyclic GMP EIA Kit (Solid Plate)** (581021.1)

Cytokines

Interleukin-1α (human) EIA Kit 583301

Stability: ≥1 year at -20°C **Limit of Detection:** 31 pg/ml

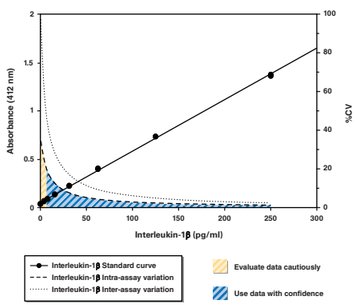
96 wells
480 wells



Interleukin-1β (human) EIA Kit 583311

Stability: ≥6 months at -20°C **Limit of Detection:** 3.9 pg/ml

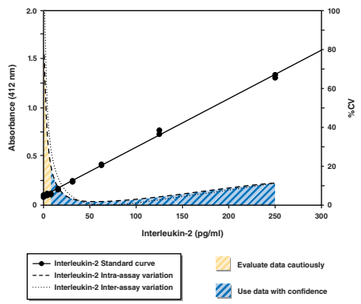
96 wells
480 wells



Interleukin-2 (human) EIA Kit 583321

Stability: ≥6 months at -20°C **Limit of Detection:** 15.6 pg/ml

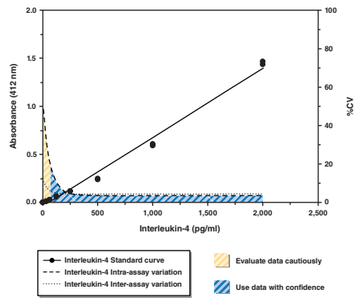
96 wells
480 wells



Interleukin-4 (human) EIA Kit 583341

Stability: ≥6 months at -20°C **Limit of Detection:** 15 pg/ml

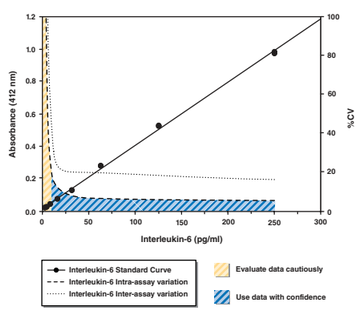
96 wells
480 wells



Interleukin-6 (human) EIA Kit 583361

Stability: ≥6 months at -20°C **Limit of Detection:** 7.8 pg/ml

96 wells
480 wells



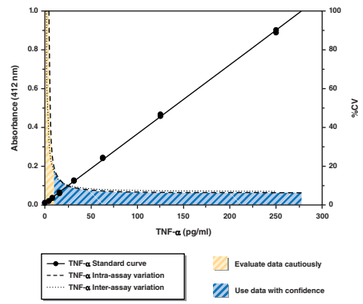
Luminex® Prostaglandin E_2/ Interleukin-1β Duplex Kit 10009597

Please see the **Prostaglandins** section for full listing on page 44

TNF-α (human) EIA Kit 589201

Stability: ≥1 year at -20°C **Limit of Detection:** 3.9 pg/ml

96 wells
480 wells



Endocrinology and Metabolism

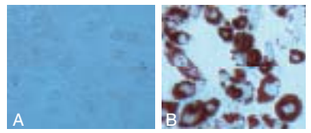
AcSDKP EIA Kit+ 589451

Please see the **Hypertension** section for full listing on page 23

Adipogenesis Assay Kit 10006908

Stability: ≥1 year at -20°C
Summary: Cayman's Adipogenesis Assay provides the reagents required for studying the induction and inhibition of adipogenesis in the established 3T3-L1 model. This kit can also be used to screen drug candidates involved in adipogenesis. The classic Oil Red O staining for lipid droplets is used in this kit as an indicator of the degree of adipogenesis, and can be quantified with a plate reader after the dye is conveniently extracted from the lipid droplet.

1 ea

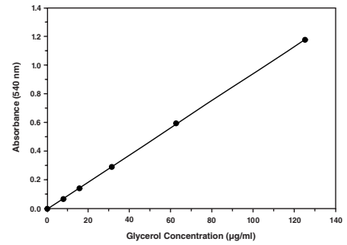


Left Panel: Non-differentiated 3T3-L1 cells. *Right Panel:* Preadipocytes differentiated for four days show lipid droplet accumulation.

Adipolysis Assay Kit 10009381

Stability: ≥1 year at -20°C
Summary: Cayman's Adipolysis Assay provides an easy to use tool for studying the hydrolysis of triglycerides to free fatty acids (FFA) and glycerol in differentiated 3T3-L1 cells. This kit will allow investigators to screen compounds involved in lipid storage and metabolism. Isoproterenol is included in the kit as a positive control for screening pharmaceuticals that regulate FFA release from adipocytes.

1 ea



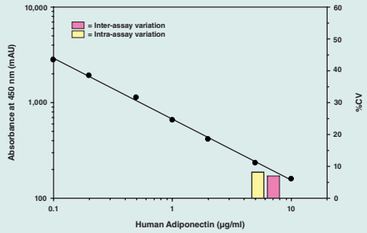
Adiponectin

Adiponectin, also known as Acpr30 and GPB-28, is a physiologically active protein which is specifically and highly expressed from adipose cells. Adipose tissue-expressed levels of adiponectin are inversely related to the degree of obesity and are correlated with insulin resistant states such as those found in obesity and type II diabetes mellitus. Adiponectin increases insulin sensitivity and decreases plasma glucose by increasing fat oxidation. The assay kits listed below are sensitive methods for the quantification of adiponectin from human or murine samples.

Adiponectin (human) EIA Kit+ 500641

Acpr30, AdipoQ, GPB-28
Stability: ≥6 months at 4°C **Limit of Detection:** 0.7 µg/ml
Summary: This EIA is based on the competition between free adiponectin and adiponectin coated to the wells of a 96-well plate for a fixed quantity of HRP-labeled adiponectin antibody.

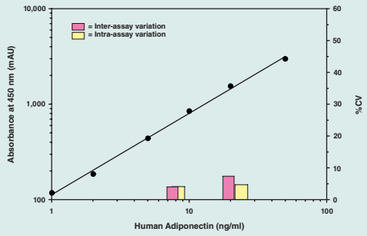
96 wells



Adiponectin (human) EIA Kit (HS)+ 10007619

Acpr30, AdipoQ, GPB-28
Stability: ≥6 months at 4°C **Limit of Detection:** 0.5 ng/ml
Summary: This EIA is based on a double antibody sandwich technique that is applicable to the quantification of both low molecular weight and high molecular weight polymers of adiponectin, but not adiponectin trimers.

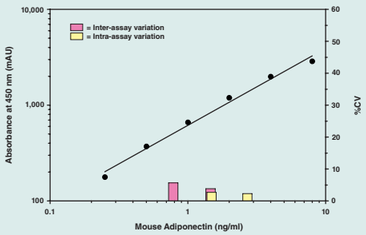
96 wells



Adiponectin (murine) EIA Kit+ 10007620

Acpr30, AdipoQ, GPB-28
Stability: ≥6 months at 4°C **Limit of Detection:** 0.1 ng/ml
Summary: This EIA is based on a double-antibody sandwich technique which utilizes a murine adiponectin-specific monoclonal capture antibody and a HRP-conjugated polyclonal antibody for detection.

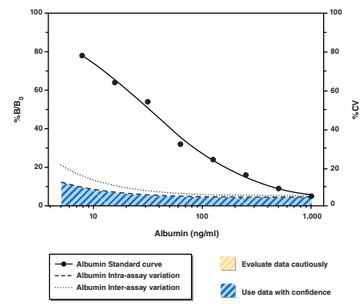
96 wells



Albumin (rat) EIA Kit+ 589801

Stability: ≥6 months at -20°C
Sensitivity: 50% B/B_0: 35 ng/ml • 80% B/B_0: 8 ng/ml
Summaary: Albumin (rat) EIA is a competitive assay that can be used for quantification of albumin in rat urine samples.

96 wells



Aldosterone EIA Kit - Monoclonal 10004377

Please see the **Steroids** section for full listing on page 52

Angiotensin II EIA Kit+ 589301

Please see the **Hypertension** section for full listing on page 23

SPI-BIO products are available through Cayman Chemical only within North & South America and Asia; elsewhere contact SPI-BIO.

Arginine Vasopressin EIA Kit 583951

Please see the **Hypertension** section for full listing on page 24

Atriopeptin (rat) EIA Kit+ 589401

Please see the **Hypertension** section for full listing on page 24

Corticosterone EIA Kit 500655

Please see the **Steroids** section for full listing on page 52

Cortisol EIA Kit 500360

Please see the **Steroids** section for full listing on page 53

Cortisol Express EIA Kit 10006791

Please see the **Steroids** section for full listing on page 53

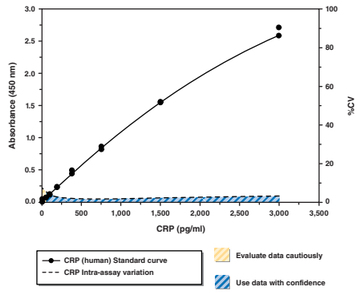
C-Reactive Protein (human) EIA Kit 10011236

CRP

Stability: ≥6 months at 4°C **Limit of Detection:** 50 pg/ml

Summary: CRP is a 224 amino acid protein that is synthesized primarily by hepatocytes, and to a lesser extent adipocytes. CRP plasma levels increase ~1,000-fold in response to acute and chronic inflammatory conditions, making it a useful gauge of inflammation in a wide range of physiological and pathological conditions. Cayman's CRP (human) EIA is a sensitive immunometric assay which can be used to measure CRP in plasma without prior sample purification.

96 wells



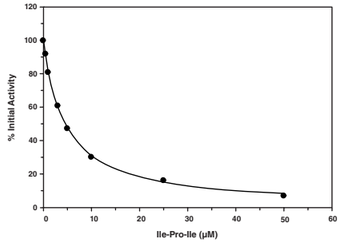
DPP (IV) Inhibitor Screening Assay Kit 700210

Dipeptidyl Peptidase (IV)

Stability: ≥6 months at -80°C

Summaary: DPP (IV) inhibitors have emerged as a new class of oral antidiabetic agents. These inhibitors promote glucose homeostasis by inhibiting degradation of glucose-dependent insulinotropic polypeptide (GIP) and glucagon-like peptide-1 (GLP-1) by DPP (IV). GLP-1 extends the action of insulin while suppressing the release of glucagon. Cayman's DPP (IV) Inhibitor Screening Assay provides a fluorescence-based method for screening DPP (IV) inhibitors in a 96-well format.

96 wells



Estradiol EIA Kit 582251

Please see the **Steroids** section for full listing on page 53

Estriol EIA Kit 582281

Please see the **Steroids** section for full listing on page 53

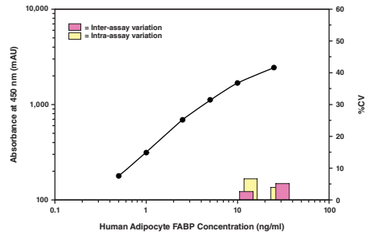
FABP4 (human) EIA Kit+ 10007614

Adipocyte-FABP, A-FABP, aP2

Stability: ≥6 months at 4°C **Limit of Detection:** 0.1 ng/ml

Summary: FAPB4 is a 15 kDa member of the intracellular FABP family, which is known for the ability to bind fatty acids and related compounds (bile acids or retinoids). FABP4 is expressed in a differentiation-dependent fashion in adipocytes and is a critical gene in the regulation of the biological function of these cells. This EIA uses a plate coated with a goat polyclonal antibody against human FABP4. Detection of bound FABP4 is achieved with a biotin-labeled anti-human FABP4 polyclonal and streptavidin-HRP.

96 wells



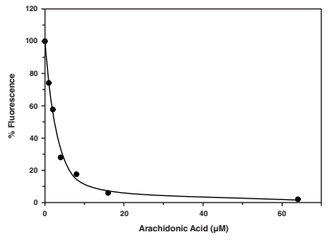
FABP4 Inhibitor/Ligand Screening Assay Kit 10010231

Adipocyte-FABP, A-FABP, aP2

Stability: ≥1 year at -80°C

Summary: Cayman's FABP4 Ligand Binding Assay provides a simple, reproducible, and sensitive tool for the identification of FABP4 ligands. The assay makes use of a detection reagent that exhibits increased fluorescence when bound to FABP4. Any strong ligand and/or inhibitor of FABP4 will displace the detection reagent thereby reducing the fluorescence.

1 ea



SPI-BIO products are available through Cayman Chemical only within North & South America and Asia; elsewhere contact SPI-BIO.

Ghrelin

Ghrelin, an endogenous ligand for the growth hormone secretagogue receptor, is synthesized principally in the stomach. It stimulates food intake and transduces signals to the hypothalamic regulatory nuclei that control energy homeostasis. The peptide consists of 28 amino acids with an octanoylation site at the serine-3 residue. Ghrelin is present in the peripheral circulation in acylated (octanoylated) and non-acylated forms in which the acylated form is biologically active. All of the following kits are based on a double-antibody sandwich technique designed to measure either the acylated or non-acylated forms of the peptide.

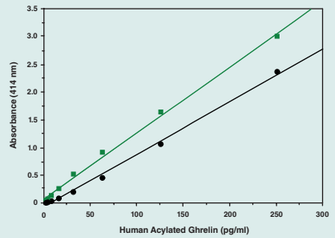
Ghrelin (human acylated) EIA Kit+ 10006306

Stability: ≥6 months at -20°C

Limit of Detection: 1.5 pg/ml after 20 hour immunological incubation
4 pg/ml after 3 hour immunological incubation

Summary: This EIA kit specifically measures the acylated form of ghrelin.

96 wells



•Also Available: **Ghrelin (human unacylated) EIA Kit+** (10008952)

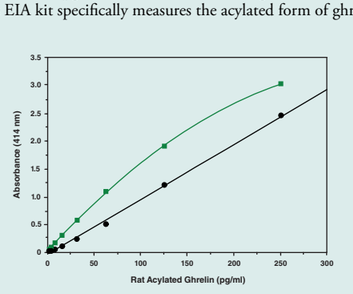
Ghrelin (rat acylated) EIA Kit+ 10006307

Stability: ≥6 months at -20°C

Limit of Detection: 1 pg/ml after 20 hour immunological incubation
3.5 pg/ml after 3 hour immunological incubation

Summary: This EIA kit specifically measures the acylated form of ghrelin.

96 wells



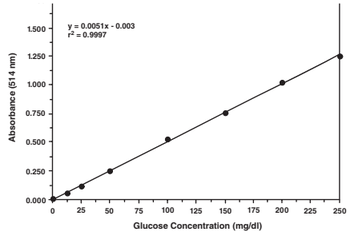
•Also Available: **Ghrelin (rat unacylated) EIA Kit+** (10008953)

Glucose Assay Kit 10009582

Stability: ≥6 months at -20°C

Summary: Cayman's Glucose Assay provides a simple, reproducible, and sensitive tool for measuring glucose in plasma, serum, and urine. In this assay, glucose is oxidized to δ-gluconolactone with concomitant reduction of the FAD-dependent enzyme glucose oxidase. Reoxidation of glucose oxidase by molecular oxygen produces H_2O_2, which is detected in a reaction resulting in formation of pink dye with an absorbance maximum at 514 nm.

192 wells



Glycerol Cell-Based Assay Kit 10011725

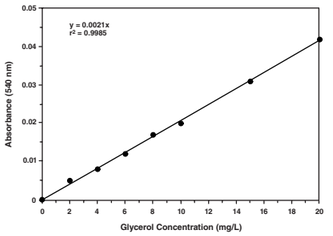
Please see the **Cell-Based Assays** section for full listing on page 8

Glycerol Assay Kit 10010755

Stability: ≥6 months at -20°C

Summary: Glycerol is the backbone of triglycerides and an important intermediate in energy metabolism being involved in both oxidative and synthetic processes. The circulating levels of glycerol and free fatty acids are considered to reflect lipolysis and are therefore useful parameters to evaluate in various conditions. Cayman's Glycerol Assay provides a simple, reproducible, and sensitive tool for measurement of glycerol in plasma and serum. The assay employs a coupled enzymatic reaction system that yields a brilliant purple product with an absorbance maximum at 540 nm.

96 wells



Growth Hormone (rat) EIA Kit+ 589601

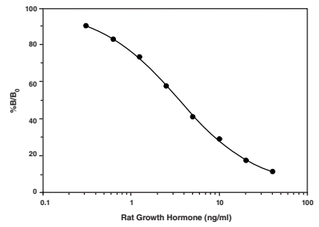
GH

Stability: ≥6 months at -20°C

Sensitivity: 50% B/B_0: 3.6 ng/ml • 80% B/B_0: 1 ng/ml

Summary: GH is a polypeptide hormone with a molecular weight of 23 kDa released from somatotropes of the anterior pituitary. It is regulated by several neurotransmitters and neuropeptides. Among other functions it plays an essential role in regulating body growth.

96 wells



Histamine EIA Kit+ 589651

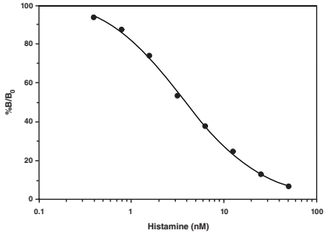
1H-imidazole 4-ethaneamine

Stability: ≥6 months at -20°C

Sensitivity: 50% B/B_0: 3.8 nM • 80% B/B_0: 1 nM

Summary: Cayman's Histamine EIA is a derivitization-amplified competitive enzyme immunoassay which detects histamine within the range from 40 to 5,500 pg/ml. The assay can be used for the analysis of histamine in blood (plasma or serum) without extraction or purification. The use of other sample types may require further processing or purification of the sample.

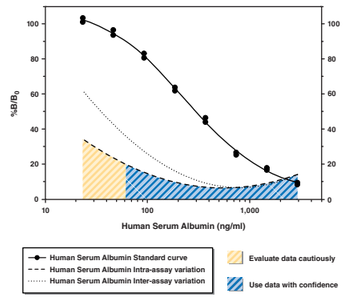
96 wells



Human Serum Albumin EIA Kit 500340

Albumin (human serum), HSA
Stability: ≥6 months at -20°C
Sensitivity: 50% B/B_0: 250 ng/ml • 80% B/B_0: 65 ng/ml
Summary: The measurement of albumin in urine is well established clinically as an indicator of renal function. HSA has many important physiological functions including maintaining osmotic pressure, buffering pH of the blood, and the transport of hormones, fatty acids, and other molecules throughout the body. Cayman's HSA EIA is a competitive assay that can be used for quantification of HSA in urine.

96 wells
480 wells

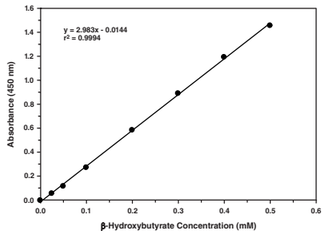


•Also Available: **Human Serum Albumin EIA Kit (Solid Plate)** (500341)

β-Hydroxybutyrate (Ketone Body) Assay Kit 700190

β-HB, 3-Hydroxybutric Acid
Stability: ≥6 months at -20°C
Summary: β-HB is a 'ketone body' which is produced in the liver, mainly from the oxidation of fatty acids, and is exported to peripheral tissues for use as an energy source. Normal ketosis can indicate that lipid metabolism has been activated and the pathway of lipid degradation is intact. Cayman's β-HB (Ketone Body) Assay provides a simple, reproducible, and sensitive tool for measuring β-HB levels in plasma, serum, or urine in a 96-well plate format with a colorimetric readout at 445-455 nm.

96 wells

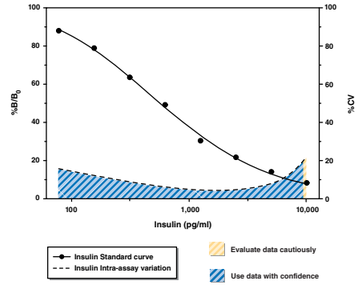


SPI-BIO products are available through Cayman Chemical only within North & South America and Asia; elsewhere contact SPI-BIO.

Insulin (rat) EIA Kit+ 589501

Stability: ≥6 months at -20°C
Sensitivity: 50% B/B_0: 0.63 ng/ml
Summary: Insulin is a polypeptide hormone synthesized by the β-cells of the islets of Langherans of the pancreas. Insulin's best known action is to lower the blood glucose concentration by increasing the rate at which glucose is converted to glycogen in the liver and muscle, and to fat in adipose tissue, by stimulating the rate of glucose metabolism, and by depressing gluconeogenesis.

96 wells



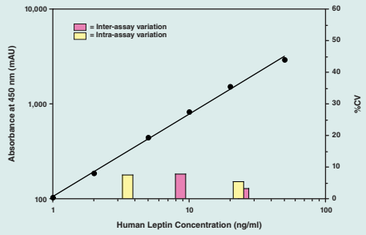
Leptin

Leptin is a 16 kDa protein hormone encoded by the obese (*ob*) gene with important effects in metabolism and regulation of body weight. Leptin has dual actions, decreasing appetite and increasing energy consumption. The primary effect of leptin appears to be mediated by leptin receptors expressed mainly in the hypothalamus. Mutations in the *ob* gene or leptin receptor gene causes hyperphagia, reduced energy expenditure, and severe obesity. The assays listed below are based on a double-antibody sandwich technique for sensitive measurement of leptin or leptin receptor.

Leptin (human) EIA Kit+ 500010

Stability: ≥6 months at 4°C **Limit of Detection:** 0.5 ng/ml

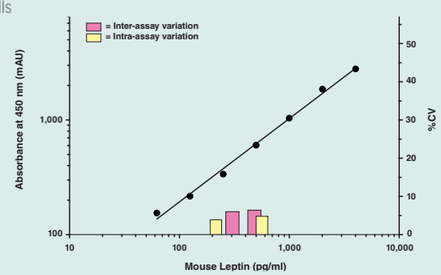
96 wells



Leptin (murine/rat) EIA Kit+ 10007609

Stability: ≥6 months at 4°C **Limit of Detection:** 50 pg/ml
Summary: This EIA utilizes plates coated with a polyclonal antibody specific for murine/rat leptin. A biotin-labeled polyclonal antibody and streptavidin-horseradish peroxidase are used for detection.

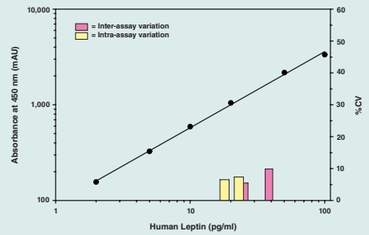
96 wells



Leptin Receptor (human) EIA Kit+ 10007608

Stability: ≥6 months at 4°C **Limit of Detection:** 0.4 ng/ml
Summary: The assay utilizes plates coated with a monoclonal capture antibody specific for the human leptin receptor and a HRP-conjugated monoclonal antibody for detection.

96 wells



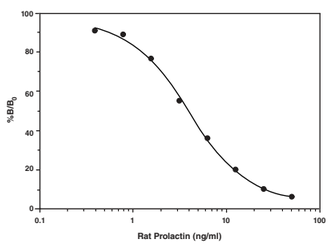
Progesterone EIA Kit 582601

Please see the **Steroids** section for full listing on page 54

Prolactin (rat) EIA Kit+ 589701

Stability: ≥6 months at -20°C
Sensitivity: 50% B/B_0: 2.8 ng/ml • 80% B/B_0: 0.9 ng/ml
Summary: Prolactin is a 199 amino acid (23 kDa) neuropeptide best known for its role in the initiation and maintenance of lactation. The hormone is secreted by the lactotrophic cells of the anterior pituitary. Numerous other biological roles of prolactin, in areas such as water and electrolyte balance, growth and development, metabolism, behavior, and immunoregulation, have been described.

96 wells



Renin Inhibitor Screening Assay Kit 10006270

Please see the **Hypertension** section for full listing on page 24

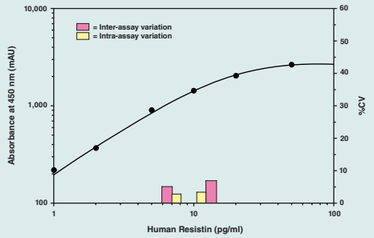
Resistin

Resistin is a peptide hormone belonging to the class of cysteine-rich secreted proteins termed the RELM family, and is also described as adipose tissue-specific secretory factor (ADSF) and Found in Inflammatory Zone (FIZZ3). Resistin impairs glucose tolerance and insulin action in mice and also inhibits adipogenesis of murine 3T3-L1 cells. Therefore, resistin has been proposed as an adipocyte secreted factor linking obesity and type 2 diabetes.

Resistin (human) EIA Kit+ 10007610

ADSF, FIZZ3
Stability: ≥6 months at 4°C **Limit of Detection:** 0.1 ng/ml
Summary: This EIA is based on a double-antibody sandwich technique for quantification of human resistin.

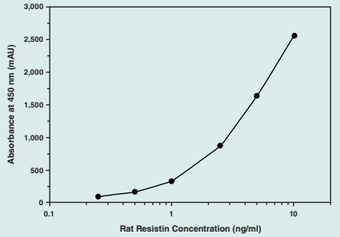
96 wells



Resistin (rat) EIA Kit+ 10007612

ADSF, FIZZ3
Stability: ≥6 months at 4°C **Limit of Detection:** 0.05 ng/ml
Summary: This EIA is based on a double-antibody sandwich technique for quantification of rat resistin.

96 wells



Testosterone EIA Kit 582701

Please see the **Steroids** section for full listing on page 54

11-keto Testosterone EIA Kit 582751

Please see the **Steroids** section for full listing on page 54

Vitellogenin Kits

Endocrine disruption is a term which refers to the harm caused by incidental exposure to environmental pesticides, industrial chemicals, or synthetic hormones which specifically target and interact with the normal endocrine hormonal system of the exposed organism. Detection of the egg yolk precursor vitellogenin (Vtg) in blood and tissue samples of juvenile and male fish is a simple and sensitive biomarker for endocrine disrupting chemicals (EDCs) and has become an accepted routine screening test for estrogenic and anti-androgenic effects of EDCs in fish. The reason for some of the success of Vtg measurements must be accredited to the sensitivity and specificity of the protein induction in fish. Importantly, the induction of Vtg is a direct physiological response to an exposure, rather than the mere presence of a chemical, and will thus take into account both bioavailability and the complex toxicokinetics and toxicodynamics that occur in animals. In collaboration with Biosense Laboratories, AS, Cayman offers a broad line of assays and antibodies for use in the detection of environmental endocrine disruptors.

Semi-Quantitative Biomarker EIA Component Kit (anti-mouse)† 10007680

Stability: ≥3 months at 4°C
Summary: This assay contains a set of reagents to be used together with a suitable Biosense primary antibody for semi-quantitative detection of biomarkers such as Vtg, CYP1A, Zrp, and MT in samples from fish.

96 wells
480 wells

Semi-Quantitative Biomarker EIA Component Kit (anti-rabbit)† 10008659

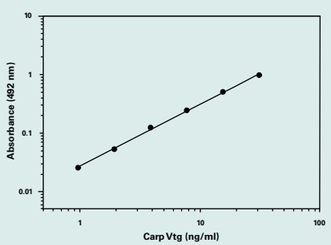
Stability: ≥3 months at 4°C
Summary: This assay contains a set of reagents to be used together with a suitable Biosense primary antibody for semi-quantitative detection of biomarkers such as Vtg, CYP1A, Zrp, and MT in samples from fish.

96 wells
480 wells

Vitellogenin (carp) EIA Kit† 10004993

Stability: ≥3 months at 4°C

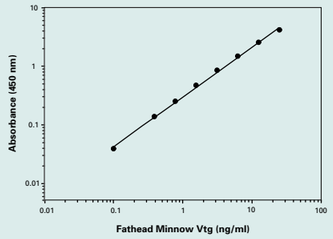
96 wells
480 wells



Vitellogenin (fathead minnow) EIA Kit† 10006943

Stability: ≥3 months at 4°C

96 wells
480 wells



Vitellogenin (medaka) EIA Kit† 10009223

Stability: ≥3 months at 4°C

96 wells
480 wells

Vitellogenin (rainbow trout) EIA Kit† 10004994

Stability: ≥3 months at 4°C

96 wells
480 wells

Vitellogenin (salmonid) Semi-Quantitative EIA Kit† 10009272

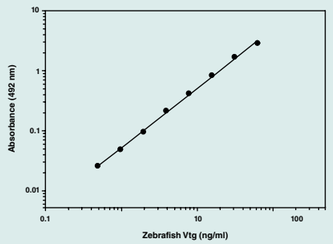
Stability: ≥3 months at 4°C

96 wells
480 wells

Vitellogenin (zebrafish) EIA Kit† 10004995

Stability: ≥3 months at 4°C

96 wells
480 wells



Biosense products are available through Cayman Chemical only within North & South America and Asia; elsewhere contact Biosense.

Enzyme Activity Kits

Cat. No.	Product Name	See Page
700260	Alanine Transaminase Activity Assay Kit	34
707002	Catalase Assay Kit	38
760151	COX Activity Assay Kit	48
700200	COX Fluorescent Activity Assay Kit	48
703102	Glutathione Peroxidase Assay Kit	39
703202	Glutathione Reductase Assay Kit	39
703302	Glutathione S-Transferase Assay Kit	39
10011563	HDAC Activity Assay Kit	12
700140	Methyltransferase Colorimetric Assay Kit	13
700150	Methyltransferase Fluorometric Assay Kit	14
10006438	Myeloperoxidase Chlorination Assay Kit	41
700160	Myeloperoxidase Peroxidation Assay Kit	41
781001	NOS Activity Assay Kit	37
760901	PAF Acetylhydrolase Assay Kit	29
765021	cPLA$_2$ Assay Kit	30
765001	sPLA$_2$ Assay Kit	30
10008041	20S Proteasome Assay Kit	10
10007892	Thioredoxin Reductase Assay Kit	43

High Throughput Screening Kits

Cat. No.	Product Name	See Page
600270	CYP3A4 Induction STEP Reporter Assay Kit (Luminescence)	51
600240	Orexin 1 Receptor STEP Reporter Assay Kit (Luminescence)	52
600250	Orexin 1 Receptor STEP Reporter Assay Kit (Luminescence)	52
10007685	PPARγ FP-Based Ligand Screening Assay Kit - Green	59
600007	Prostaglandin D Synthase (hematopoietic-type) FP-Based Inhibitor Screening Assay Kit - Green	44
500581	Prostaglandin D$_2$ FPIA Kit - Green	45
10007835	Prostaglandin D$_2$ FPIA Kit - Red	45
500501	Prostaglandin E$_2$ FPIA Kit - Green	49
10004517	Prostaglandin E$_2$ FPIA Kit - Red	49

Cayman supplies several focused libraries. Please see the Chemical Library section on page 11.

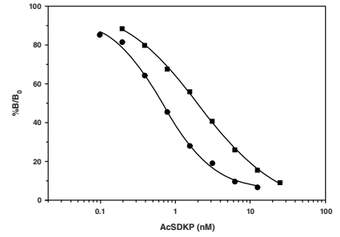
SPI-BIO products are available through Cayman Chemical only within North & South America and Asia; elsewhere contact SPI-BIO.

Hypertension

AcSDKP EIA Kit+ 589451

N-Acetyl Ser-Asp-Lys-Pro
Stability: ≥6 months at -20°C
Sensitivity: 50% B/B$_0$: 2.0 nM after 3 hour immunological reaction
0.5 nM after 18 hour immunological reaction
80% B/B$_0$: 0.2 nM after 18 hour immunological reaction
Summary: AcSDKP is a tetrapeptide growth regulatory hormone which inhibits the proliferation of hematopoietic stem cells. The dipeptidase, angiotensin converting enzyme (ACE), actively metabolizes circulating AcSDKP, giving it a brief plasma half-life of 4 to 5 minutes. ACE inhibition is a major therapeutic end point in the treatment of hypertension management. A further consequence of ACE inhibition is the accumulation of AcSDKP in plasma and urine. This accumulation may have physiological effects, which are manifested as the anemia of chronic ACE inhibitor toxicity. More commonly, plasma and urine AcSDKP levels can be used as a biomarker of ACE inhibition and an index of patient compliance with therapy. Measurement of AcSDKP in human urine or plasma can be readily accomplished by EIA.

96 wells



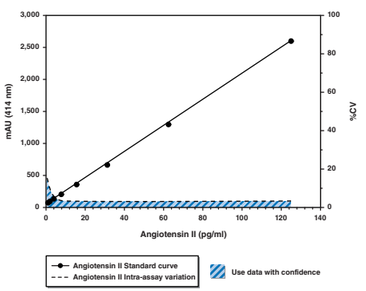
Aldosterone EIA Kit - Monoclonal 10004377

Please see the **Steroids** section for full listing on page 52

Angiotensin II EIA Kit+ 589301

Stability: ≥6 months at -20°C **Limit of Detection:** 1.5 pg/ml
Summary: Angiotensin II is a primary reactive vasoconstrictor, the main stimulus for aldosterone release, and one of the causative factors of chronic hypertension. The active angiotensin II octapeptide is released *via* a tightly controlled series of prohormones and proteases. The unique, patented 'Immobilized Antigen' technology of this angiotensin II immunometric assay allows reliable detection of 1-2 pg/ml, or as little as 10% of the normal human plasma angiotensin II concentration.

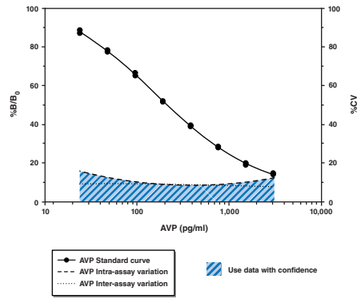
96 wells



Arginine Vasopressin EIA Kit 583951

ADH, Antidiuretic Hormone, Argipressin, AVP, Vasopressin
Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 300 pg/ml • 80% B/B_0: 50 pg/ml
Summary: AVP is a nine amino acid peptide hormone that plays a primary role in the regulation of renal water excretion and a secondary role in the regulation of cardiovascular function in mammals. Cayman's AVP EIA is a competitive assay that can be used for the measurement of AVP from plasma and serum.

96 wells
480 wells

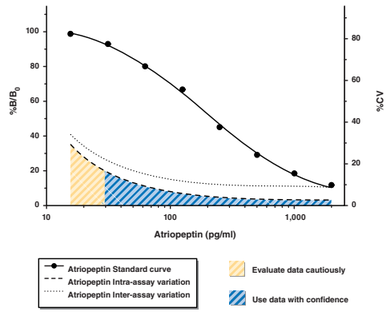


•Also Available: **Arginine Vasopressin EIA Kit (Solid Plate)** (10009345)

Atriopeptin (rat) EIA Kit+ 589401

Stability: ≥6 months at -20°C
Sensitivity: 50% B/B_0: 190 pg/ml • 80% B/B_0: 60 pg/ml
Summary: Atriopeptin is a 28 amino acid peptide synthesized primarily in cardiac atria. This peptide hormone acts in opposition to angiotensin II in regulating renal, hemodynamic, and endocrine function. Atriopeptin is released in response to the increased pressure and mechanical stretch of the right atrium due to blood volume overload. Atriopeptin then acts at the nephron to increase salt and water excretion, lowering blood volume and blood pressure.

96 wells



CGRP (human) EIA Kit+ 589101

Please see the **Bioactive Peptides** section for full listing on page 6

CGRP (rat) EIA Kit+ 589001

Please see the **Bioactive Peptides** section for full listing on page 6

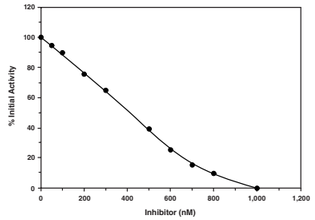
Endothelin EIA Kit 583151

Please see the **Bioactive Peptides** section for full listing on page 6

Renin Inhibitor Screening Assay Kit 10006270

Stability: ≥6 months at -80°C
Summary: Cayman's Renin Inhibitor Screening Assay provides a convenient assay in a 96-well format for evaluating human renin inhibitors. The assay utilizes a renin-based synthetic peptide substrate which incorporates the fluorophore EDANS at one end and an EDAN-quenching molecule (Dabcyl) at the other end. After cleavage by renin, the peptide-EDANS product is released yielding bright fluorescence.

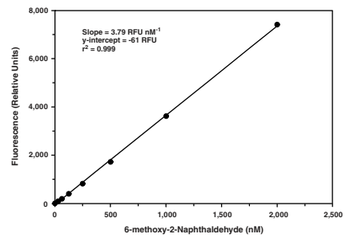
96 wells



Soluble Epoxide Hydrolase Cell-Based Assay Kit 600090

sEH
Stability: ≥1 year at -80°C
Summary: Mammalian sEH is a member of the α/β-hydrolase fold family of enzymes that catalyze the hydrolysis of exogenous and endogenous epoxides to vicinal diols. Endogenous substrates for sEH include EETs which exhibit vasodilatory and anti-inflammatory activity. Cayman's sEH Cell-Based Assay provides a fluorescence-based method for detecting epoxide hydrolase activity in whole cells. The assay utilizes Epoxy Fluor 7, a sensitive fluorescent substrate for sEH that can be used to monitor the activity of both human and murine enzymes.

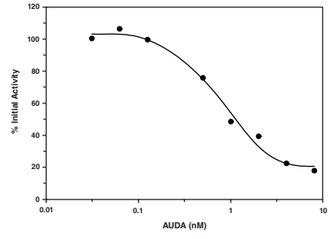
480 wells



Soluble Epoxide Hydrolase Inhibitor Screening Assay Kit 10011671

sEH
Stability: ≥6 months at -80°C
Summary: Small molecule inhibitors of sEH may hold promise as therapeutics for the treatment of hypertension and vascular inflammation. Cayman's fluorescence-based sEH Inhibitor Screening Assay provides a convenient method for screening epoxide hydrolase inhibitors. The assay utilizes (3-phenyl-oxiranyl)-acetic acid cyano-(6-methoxy-naphthalen-2-yl)-methyl ester (PHOME) as a substrate. Hydrolysis of PHOME by epoxide hydrolase produces the highly fluorescent 6-methoxy-2-naphthaldehyde.

96 wells

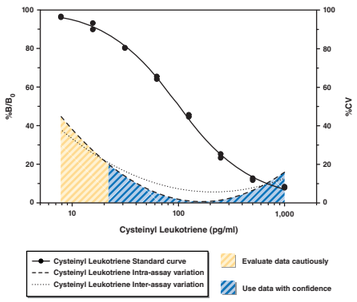


SPI-BIO products are available through Cayman Chemical only within North & South America and Asia; elsewhere contact SPI-BIO.

Leukotrienes

Cysteinyl Leukotrienes

The conjugation of glutathione to LTA_4 results in the formation LTC_4. LTC_4 is rapidly metabolized to LTD_4 and LTE_4. This metabolism is essentially complete within 10 minutes in the human lung. LTC_4, LTD_4, and LTE_4 are collectively referred to as CysLTs. LTC_4 and LTD_4 are potent mediators of asthma and hypersensitivity. They induce bronchoconstriction, increase microvascular permeability, and are vasoconstrictors of coronary arteries. CysLTs can accumulate to relatively high concentrations in the effusion fluids associated with inflammation (*e.g.*, ascites fluid, synovial fluid, pleural effusion, pericardial, or cerebral aspirates). Since LT metabolism is incomplete in these circumstances, substantial amounts of LTC_4, LTD_4, and LTE_4 may be present (*e.g.*, bronchalveolar lavage fluid from asthmatic subjects may contain 700-1,000 pg/ml of CysLTs comprised mainly of LTC_4 and LTD_4). CysLTs are excreted in urine as intact LTE_4 (~9-12%) and LTE_4 metabolites. Since LTC_4 and LTD_4 are virtually absent from urine, the CysLT measurement in urine is often best accomplished by measuring LTE_4. Cultured cells synthesizing LTC_4 will generally release it into the medium where it will accumulate without further metabolism.



Cysteinyl Leukotriene Affinity Purification Kit (4 ml) 10010392

Stability: ≥2 years at 4°C
Summary: This kit contains all reagents necessary for simple one-step purification of cysteinyl leukotrienes from most biological samples.

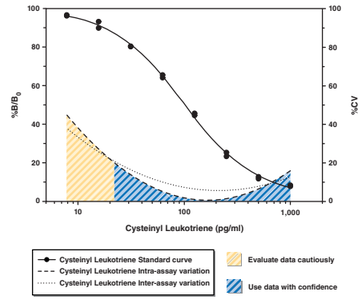
1 ea
5 ea

•Also Available: **Cysteinyl Leukotriene Affinity Column** (10005362)
Cysteinyl Leukotriene Affinity Purification Kit (20 ml) (520503)
Cysteinyl Leukotriene Affinity Sorbent (420509)

Cysteinyl Leukotriene EIA Kit 520501

Stability: ≥6 months at -80°C
Sensitivity: 50% B/B_0: 80 pg/ml • 80% B/B_0: 13 pg/ml
Summary: Cayman's CysLT EIA is a competitive assay that can be used for quantification of CysLTs in urine, culture media, and other sample matrices.

96 wells
480 wells

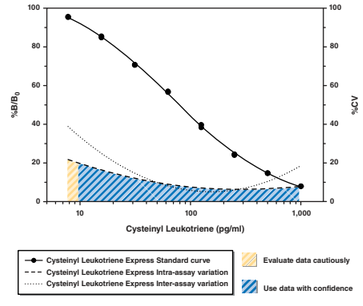


•Also Available: **Cysteinyl Leukotriene EIA Kit (Solid Plate)** (520501.1)

Cysteinyl Leukotriene Express EIA Kit 10009291

Stability: ≥6 months at -80°C
Sensitivity: 50% B/B_0: 80 pg/ml • 80% B/B_0: 20 pg/ml
Summary: Cayman's CysLT Express EIA is a competitive assay that can be used for measurement of all three primary CysLTs (LTC_4, LTD_4, and LTE_4) in urine, culture media, and other sample matrices. This sensitive assay takes about three hours to complete.

96 wells
480 wells



•Also Available: **Cysteinyl Leukotriene Express EIA Kit (Solid Plate)** (500004)

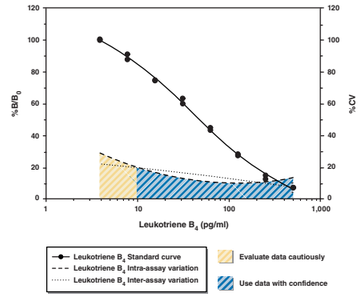
Recommendations for CysLT Kits

Sample Type:	LTC_4 EIA Kit	LTE_4 EIA Kit	CysLT EIA Kit
Plasma/Serum	Rapid metabolism of LTs; excreted in urine		
Urine	not applicable	✓	✓
Bronchiolar Lavage; effusion fluids	not recommended	not recommended	All CysLTs may be present in relatively high concentrations
Cell Culture	✓	not applicable	not applicable

Leukotriene B_4 EIA Kit — 520111

Stability: ≥6 months at -20°C
Sensitivity: 50% B/B_0: 50 pg/ml • 80% B/B_0: 13 pg/ml
Summary: LTB_4 is synthesized from arachidonic acid by the combined action of 5-LO and LTA_4 hydrolase. LTB_4 has long been recognized as a potent mediator of inflammation. It stimulates a number of leukocyte functions, including aggregation, stimulation of ion fluxes, enhancement of lysosomal enzyme release, superoxide anion production, chemotaxis, and chemokinesis. In subnanomolar ranges (3.9×10^{-10} M), LTB_4 causes chemotaxis and chemokinesis in human PMNLs.

96 wells
480 wells

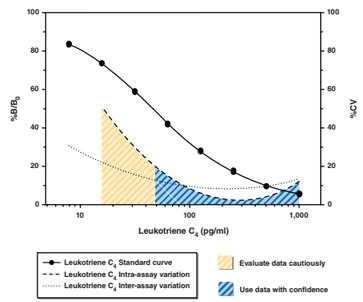


• Also Available: **Leukotriene B_4 EIA Kit (Solid Plate)** (520111.1)

Leukotriene C_4 EIA Kit — 520211

Stability: ≥6 months at -80°C
Sensitivity: 50% B/B_0: 45 pg/ml • 80% B/B_0: 10 pg/ml
Summary: Cayman's LTC_4 Assay is a competitive EIA that can best be used for the quantification of LTC_4 in select sample types. Cultured cells synthesizing LTC_4 will generally release it into the medium where it will accumulate without further metabolism. Plasma levels of LTC_4 are virtually undetectable.

96 wells
480 wells

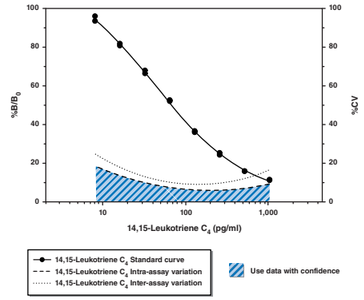


• Also Available: **Leukotriene C_4 EIA Kit (Solid Plate)** (520211.1)

14,15-Leukotriene C_4 EIA Kit — 10006748

Stability: ≥6 months at -80°C
Sensitivity: 50% B/B_0: 66 pg/ml • 80% B/B_0: 18 pg/ml
Summary: Although the majority of LTs are formed through the 5-LO pathway, a second family of LTs can be formed through an alternate pathway involving the dual actions of 15- and 12-LOs. The resulting epoxytriene, 14,15-LTA_4, can be converted to 14,15-LTC_4. Although many of the physiological actions of 14,15-LTC_4 remain to be elucidated, it has been shown to have weak contractile activity on both guinea pig ileum and pulmonary parenchyma.

96 wells
480 wells
$1260.00

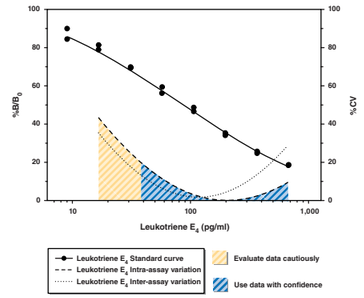


• Also Available: **14,15-Leukotriene C_4 EIA Kit (Solid Plate)** (10006749)

Leukotriene E_4 EIA Kit — 520411

Stability: ≥6 months at -80°C
Sensitivity: 50% B/B_0: 100 pg/ml • 80% B/B_0: 25 pg/ml
Summary: Cayman's LTE_4 EIA is a competitive assay that can be used for quantification of LTE_4 in urine, plasma, serum, whole blood, and other heterogeneous mixtures such as lavage fluids, aspirates, and other sample matrices.

96 wells
480 wells

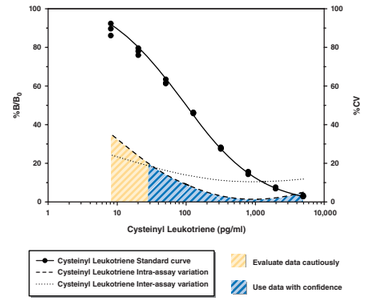


• Also Available: **Leukotriene E_4 EIA Kit (Solid Plate)** (520411.1)

Luminex® Cysteinyl Leukotriene Kit — 10007577

Stability: ≥6 months at -80°C
Sensitivity: 50% B/B_0: 90 pg/ml • 80% B/B_0: 15 pg/ml
Summary: For application of this technology to the measurement of CysLT in a competitive format, microspheres have been coated with Cayman's CysLT monoclonal antibody. The assay is based on the competition between CysLT and a CysLT-phycoerytherin conjugate (CysLT tracer) for monoclonal antibody binding sites on the microsphere beads.

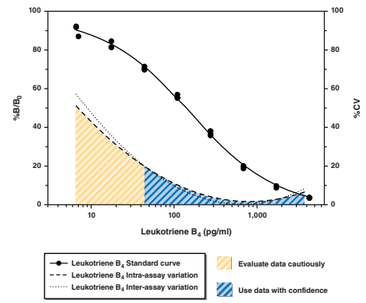
96 wells



Luminex® Leukotriene B_4 Kit — 500260

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 138 pg/ml • 80% B/B_0: 24 pg/ml
Summary: LTB_4 is a potent mediator of inflammation that stimulates a number of leukocyte functions, including aggregation, stimulation of ion fluxes, enhancement of lysosomal enzyme release, superoxide anion production, chemotaxis, and chemokinesis. Cayman's Luminex® LTB_4 can be used for quantification of LTB_4 in plasma, culture media, and other sample matrices.

1 ea



Inhibitor Screening Kits

Cat. No.	Product Name	See Page
600070	BACE Inhibitor Screening Assay Kit	37
760111	Colorimetric COX (ovine) Inhibitor Screening Assay Kit	48
700100	COX Fluorescent Inhibitor Screening Assay Kit	48
560131	COX Inhibitor Screening Assay Kit	48
560101	COX (ovine) Inhibitor Screening Assay Kit	48
10005196	FAAH Inhibitor Screening Assay Kit	28
10010231	FABP4 Inhibitor/Ligand Screening Assay Kit	18
10006515	HAT Inhibitor Screening Assay Kit	12
10011564	HDAC1 Inhibitor Screening Assay Kit	13
700230	HDAC8 Inhibitor Screening Assay Kit	13
700360	JMJD2A Inhibitor Screening Assay Kit	13
700370	JMJD2D Inhibitor Screening Assay Kit	13
760700	Lipoxygenase Inhibitor Screening Assay Kit	29
700120	LSD1 Inhibitor Screening Assay Kit	13
705192	Monoacylglycerol Lipase Inhibitor Screening Assay Kit	29
700170	Myeloperoxidase Inhibitor Screening Assay Kit	41
10004380	PAF Acetylhydrolase Inhibitor Screening Assay Kit	30
600007	Prostaglandin D Synthase (hematopoietic-type) FP-Based Inhibitor Screening Assay Kit - Green	44
10006595	Prostaglandin D Synthase Inhibitor Screening Assay Kit	44
10006270	Renin Inhibitor Screening Assay Kit	24
10010401	SIRT1 Direct Fluorescent Screening Assay Kit	14
10010991	SIRT1 FRET-Based Screening Assay Kit	15
10011566	SIRT3 Direct Fluorescent Screening Assay Kit	14
10011671	Soluble Epoxide Hydrolase Inhibitor Screening Assay Kit	24
10004883	$sPLA_2$ (Type V) Inhibitor Screening Assay Kit	31

Luminex® Assay Kits

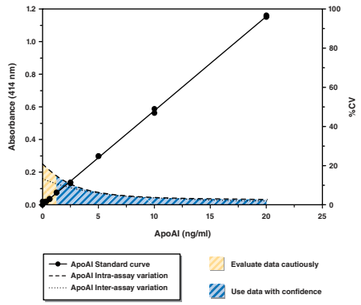
Cat. No.	Product Name	See Page
10007577	Luminex® Cysteinyl Leukotriene Kit	27
500260	Luminex® Leukotriene B_4 Kit	27
10007501	Luminex® Prostaglandin E_2 Kit	44
10009597	Luminex® Prostaglandin E_2/Interleukin-1β Duplex Kit	44
10007502	Luminex® Thromboxane B_2 Kit	55
10010971	Luminex® 11-dehydro Thromboxane B_2 Kit	55

Lipids

ApoAI (human) EIA Kit 500155

Apolipoprotein AI
Stability: ≥1 year at -20°C **Sensitivity:** 0.3 ng/ml
Summary: ApoAI is a major protein component of HDL. Clinical studies have demonstrated that lower levels of ApoAI are associated with an increased risk of myocardial infarction and coronary artery disease. Overexpression of ApoAI raises HDL cholesterol levels and inhibits the progression of atherosclerosis in mice. Cayman's ApoAI (human) EIA is an immunometric assay which can be used to accurately detect and quantify ApoAI in plasma and serum without prior sample purification.

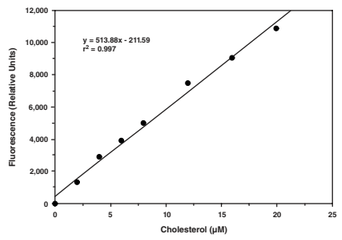
96 wells
480 wells



Cholesterol Assay Kit 10007640

Stability: ≥1 year at -20°C
Summary: Cholesterol, particularly in the form of LDLs, is well understood to be associated with increased risk of coronary heart disease. The measurement of cholesterol is one of the most common tests performed in the clinical laboratory setting. However, simple and easy assays for cholesterol in the research lab have not been readily available. The Cayman's Cholesterol Assay provides a simple fluorometric method for the sensitive quantitation of total cholesterol in plasma or serum.

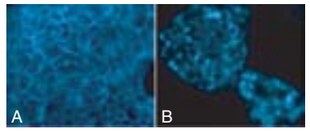
96 wells
480 wells



Cholesterol Cell-Based Detection Assay Kit 10009779

Stability: ≥6 months at -20°C **Detection Method(s):** Fluorescence microscope
Summary: The mechanism for the movement of cholesterol from intracellular sites to their ultimate cellular destination is an unresolved question of fundamental importance to cell biology and medicine. Cayman's Cholesterol Cell-based Detection Assay provides a simple fluorometric method to study mechanisms and biological factors that regulate cholesterol metabolism or movement within cells.

192 wells

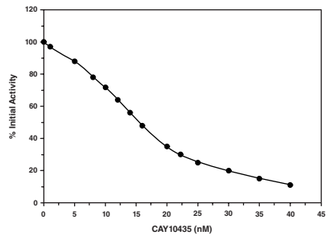


Accumulation of cholesterol in HepG2 cells. *Panel A:* Cells treated with DMSO (vehicle). *Panel B:* U-18666A (1.25 µM) treatment for 48 hours induces intracellular accumulation of cholesterol droplets.

FAAH Inhibitor Screening Assay Kit 10005196

Stability: ≥6 months at -80°C
Summary: FAAH is a cytosolic serine hydrolase responsible for the degradation of fatty acid amides, including AEA. Finding inhibitors to FAAH could offer a beneficial approach toward the treatment of pain, obesity, and various neurological diseases. Cayman's FAAH Inhibitor Screening Assay provides a fluorescence-based method for screening FAAH inhibitors, using AMC-arachidonoyl amide as the substrate.

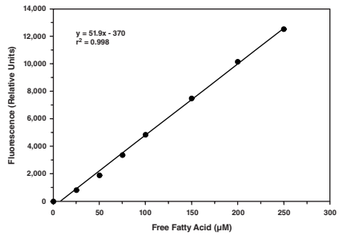
96 wells



Free Fatty Acid Assay Kit 700310

Stability: ≥6 months at -20°C
Summary: The measurement of FFA can be useful in determining metabolic status. Cayman's FFA Assay provides a simple, reproducible, and sensitive tool for measuring free fatty acids in plasma, serum, and urine. The FFA Assay utilizes a coupled enzymatic reaction that results in generation of the highly fluorescent product resorufin.

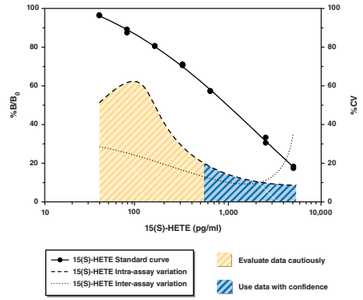
96 wells



15(S)-HETE EIA Kit 534721

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 960 pg/ml • 80% B/B_0: 170 pg/ml
Summary: 15(S)-HETE is produced from arachidonic acid by the enzyme 15-LO. In humans it is formed primarily in the respiratory epithelium, leukocytes, and reticulocytes. It may contribute to allergic rhinitis, has anti-inflammatory properties, and acts as a vasoconstrictor. Cayman's 15(S)-HETE EIA is a competitive assay that is very specific for 15(S)-HETE, showing very low cross reactivity to other HETEs.

96 wells
480 wells

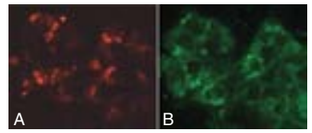


•Also Available: **15(S)-HETE EIA Kit (Solid Plate)** (534721.1)

LDL Uptake Cell-Based Assay Kit 10011125

Stability: ≥6 months at 4°C **Detection Method(s):** Fluoresence microscope
Summary: LDL uptake and its regulation are important therapeutic targets for atherosclerosis and related diseases. Cayman Chemical's LDL Uptake Cell-Based Assay employs a preparation of human LDL conjugated to Dylight™ 549 as a fluorescent probe for detection of LDL uptake into cultured cells. A LDL receptor-specific antibody and a Dylight™ 488-conjugated secondary antibody are included in the kit for identifying the distribution of LDL receptors.

1 ea

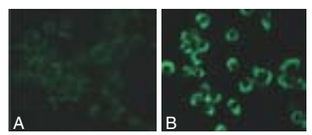


LDL Uptake in HepG2 cells. HepG2 cells were treated with 32 µM EGCG overnight followed by addition of LDL-DyLight™ 549 for four hours. *Panel A:* DyLight™ 549 taken into cells appear in red. *Panel B:* LDL receptors in green show.

Lipid Droplets Fluorescence Assay Kit 500001

Stability: ≥1 year at -20°C
Detection Method(s): FC, Fluorescence microscope, and Plate reader
Summary: Lipid droplets are a fundamental component of intracellular lipid homeostasis in all cell types and they provide a rapidly mobilized lipid source for many important biological processes. Cayman's Lipid Droplets Fluorescence Assay can be used to study regulators of lipid droplet biogenesis. The main advantage of this assay is that the green fluorescence of Nile Red is both very sensitive and specific for lipid droplets.

480 tests

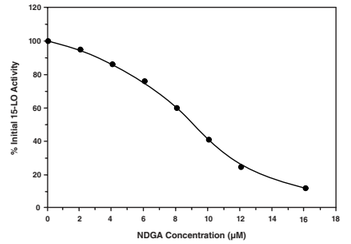


Oleic Acid dramatically induces lipid droplet accumulation in neuro-2a cells. *Left Panel:* Neuro-2a cells were treated with vehicle or 400 µM oleic acid *Right Panel:* for 24 hours and then processed for lipid droplet staining.

Lipoxygenase Inhibitor Screening Assay Kit 760700

Stability: ≥1 year at 4°C
Summary: This assay kit provides an accurate and convenient method for screening LO inhibitors. This assay measures the hydroperoxides generated from the incubation of a LO (5-, 12-, or 15-LO) with either arachidonic or linoleic acid.

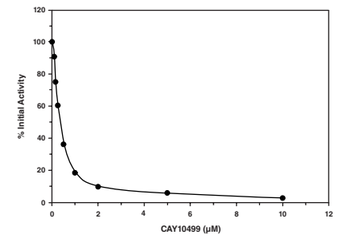
96 wells



Monoacylglycerol Lipase Inhibitor Screening Assay Kit 705192

MAGL, MGL
Stability: ≥6 months at -80°C
Summary: MAGL is the main enzyme responsible for the inactivation of 2-AG. Cayman's Monoacylglycerol Lipase Inhibitor Screening Assay provides a convenient method for screening human MAGL inhibitors. MAGL hydrolyzes 4-nitrophenylacetate resulting in a yellow product, 4-nitrophenol, with an absorbance of 405-412 nm.

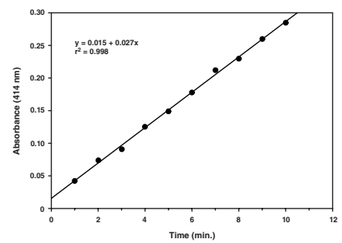
96 wells



PAF Acetylhydrolase Assay Kit 760901

Lp-PLA$_2$, PAF-AH
Stability: ≥1 year at -20°C
Summary: PAF-AH catalyzes the hydrolysis of the potent biologically-active phospholipid PAF, generating inactive lyso-PAF. Cayman's PAF-AH assay kit provides an accurate and convenient method for measurement of PAF-AH activity. The assay uses 2-thio PAF which serves as a substrate for PAF-AH. Upon hydrolysis of the acetyl thioester bond at the *sn*-2 position by PAF-AH, free thiols are detected using Ellman's reagent.

96 wells

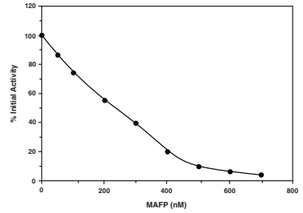


PAF Acetylhydrolase Inhibitor

Screening Assay Kit 10004380

Lp-PLA$_2$, PAF-AH
Stability: ≥6 months at -20°C
Summary: Cayman's PAF-AH Inhibitor Screening Assay uses 2-thio PAF as a substrate for PAF-AH. Upon hydrolysis of the acetyl thioester bond at the *sn*-2 position by PAF-AH, free thiols are detected using DTNB (Ellman's reagent).

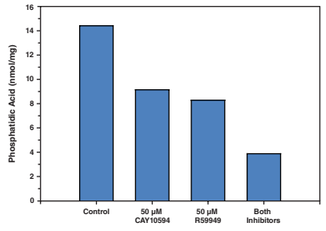
96 wells



Phosphatidic Acid Assay Kit 700240

PA
Stability: ≥6 months at -20°C
Summary: PA is a common phospholipid that is a major constituent of cell membranes and a central intermediate for the synthesis of membrane lipids and storage lipids. Cayman's Phosphatidic Acid Assay provides a fluorescence-based method for measuring PA. Lipase is used to hydrolyze PA in cellular lipids to glycerol-3-phosphate. A coupled enzyme reaction system then generates the highly fluorescent compound resorufin (excitation 530-540 nm and emission 585-595 nm).

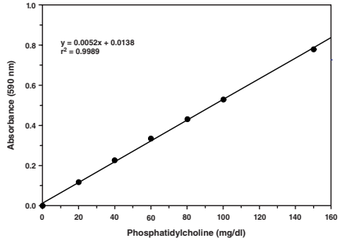
96 wells



Phosphatidylcholine Assay Kit 10009926

PC
Stability: ≥6 months at -20°C
Summary: Cayman's PC Assay provides a specific, sensitive, and convenient method for quantifying PC in plasma or serum. In this assay, PC-specific PLD hydrolyzes PC to choline and PA. A couple enzyme reaction system utilizes the choline and generates a blue dye with maximum absorption at 595 nm.

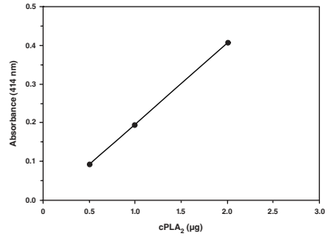
96 wells



cPLA$_2$ Assay Kit 765021

PLA$_2$ Type IV
Stability: ≥1 year at -20°C
Summary: Arachidonoyl thio-PC is a substrate for cPLA$_2$ by virtue of the presence of arachidonic acid at the *sn*-2 position of the glycerophospholipid. Hydrolysis of the arachidonoyl thioester bond at the *sn*-2 position by PLA$_2$ releases free thiol which can be detected by DTNB. This assay can be used to determine the activity of cPLA$_2$ in purified preparations, cell cultures, or tissue homogenates that are known to contain only cPLA$_2$. Use of this assay with preparations containing more than one type of PLA$_2$ will result in the measurement of total PLA$_2$ activity rather than cPLA$_2$ alone. Isozyme-specific cPLA$_2$ activity can be measured by excluding sPLA$_2$ or inhibiting iPLA$_2$ activities in the assay.

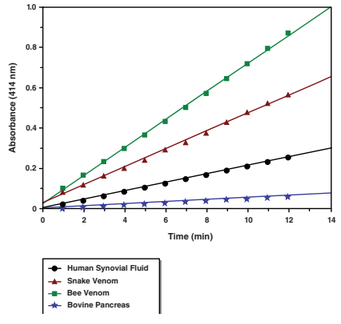
96 wells



sPLA$_2$ Assay Kit 765001

Secretory PLA$_2$ (Type IIA)
Stability: ≥1 year at -20°C
Summary: Cayman's sPLA$_2$ Assay provides an accurate and convenient method for measurement of sPLA$_2$ activity. This assay uses the 1,2-dithio analog of diheptanoyl phosphatidylcholine which serves as a substrate for most PLA$_2$s with the exception of cPLA$_2$. Upon hydrolysis of the thioester bond at the *sn*-2 position by PLA$_2$, free thiols are detected using DTNB.

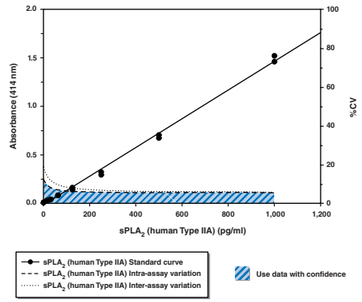
96 wells



sPLA$_2$ (human Type IIA) EIA Kit 585000

SPLA$_2$ (human synovial)
Stability: ≥1 year at -20°C **Limit of Detection:** 15 pg/ml
Summary: Cayman's sPLA$_2$ EIA is an immunometric (*i.e.* "sandwich") assay that can be used for the quantification of sPLA$_2$ in plasma, synovial fluid, and other sample matrices.

96 wells
480 wells

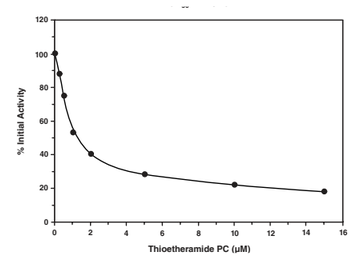


- Also Available: **sPLA$_2$ (human Type IIA) Affinity Sorbent** (485009)

sPLA$_2$ (Type V) Inhibitor Screening Assay Kit 10004883

SPLA$_2$ (Type V)
Stability: ≥1 year at -20°C
Summary: The sPLA$_2$ (Type V) Inhibitor Screening Assay is a colorimetric assay designed for rapid screening of Type V sPLA$_2$ inhibitors in a 96-well format.

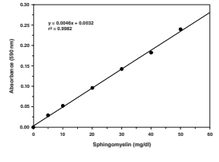
96 wells



Sphingomyelin Assay Kit 10009928

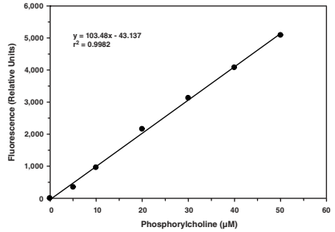
SM
Stability: ≥6 months at 4°C
Summary: Sphingomyelin is an important lipid component of cell membranes and lipoproteins. Cayman's SM Assay provides a specific and sensitive method for quantifying SM in plasma or serum in a 96-well format with a colorimetric readout at 595 nm.

96 wells



Sphingomyelinase Assay Kit 10006964

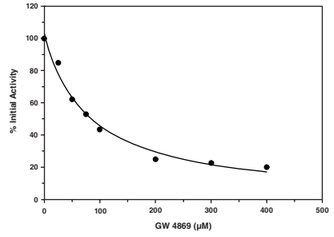
SMase
Stability: ≥6 months at -20°C
Summary: Cayman's Sphingomyelinase Assay provides a simple, reproducible, and sensitive tool for measuring neutral and acidic sphingomyelinase activity from tissue homogenates, cell lysates, serum, saliva, and urine. The assay employs a coupled enzymatic reaction system, resulting in the formation of highly fluorescent resorufin.



Sphingomyelinase Inhibitor Screening Assay Kit 700330

SMase
Stability: ≥6 months at -20°C
Summary: Cayman's Sphingomyelinase Inhibitor Screening Assay is a novel method for screening sphingomyelinase inhibitors. Cleavage of a unique sphingomyelin conjugate by SMase in the assay results in the release of a ceramide analog containing a free thiol which is detected by the SMase Detector to yield a highly fluorescent product.

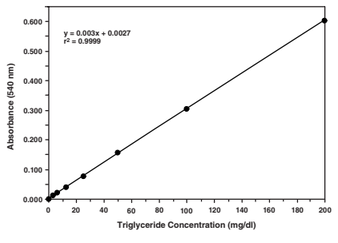
96 wells



Triglyceride Assay Kit 10010303

TG
Stability: ≥6 months at -20°C
Summary: The measurement of TG levels are useful in the diagnosis of primary and secondary hyperlipoproteinemia, dyslipidemia, and triglyceridemia. Cayman's TG Assay provides a simple, reproducible, and sensitive tool for measuring TGs in plasma and serum. The assay is initiated with the enzymatic hydrolysis of the TGs by lipase to produce glycerol and free fatty acids. The glycerol released is subsequently measured by a coupled enzymatic reaction system with a colorimetric readout at 540 nm.

96 wells



vol. 9 Ak

Tom Brock, Ph.D.

Oxidative Stress Assay Kits

Oxidative stress is an ongoing battle between highly reactive free radicals and the systems designed to mitigate their effects. When free radicals are winning this battle, oxidative damage occurs, initiating repair and removal processes. In a biological system like the human body, oxidative stress may be seen to start at the molecular level, with a direct interaction between free radicals and a protein, a lipid, carbohydrates, or nucleic acids. Certain diseases might stem from a failure to counter these effects, with damage spreading locally or, perhaps, systemically. Alternatively, oxidative damage can result from broad oxidant production throughout a tissue or organ that simply overwhelms a functional antioxidant program. Detection and evaluation of these processes can be pursued in many ways, as reflected by the numerous assay kits offered by Cayman. This section presents some of those kits and their uses.

Screening Assays

Certain kits are intended to be used primarily for answering the general question: Is there a change in oxidative stress under certain conditions? They provide little information regarding where or when the stress occurred. For example, the measurement of 'Thiobarbituric Acid Reactive Substances' (**TBARS Assay Kit, Catalog No. 10009055**) is a well-established method for screening and monitoring lipid peroxidation. TBA reacts with malondialdehyde, which is derived from lipid hydroperoxides produced by oxidative stress. This kit provides a simple, reproducible, and standardized tool for assaying lipid peroxidation in plasma, serum, urine, tissue homogenates, and cell lysates. A positive result from the TBARS analysis supports further investigation, for example, into the causes, consequences, or prevention of the detected lipid peroxidation.

Similarly, the **Hydrogen Peroxide (urinary) Assay Kit (Catalog No. 706011)** can be used to detect changes in secreted hydrogen peroxide (H_2O_2) as part of a preliminary study. This simple, sensitive, and specific assay allows the researcher to monitor a metabolic indicator of oxidative stress (H_2O_2) non-invasively by testing urine samples. Again, a detected increase in urinary H_2O_2 provides the rationale for more detailed studies.

A versatile screening assay is provided by the **Antioxidant Assay Kit (Catalog No. 709001)**. The overall antioxidant capacity may give more relevant biological information compared to that obtained by the measurement of individual components, since it considers the cumulative effect of all antioxidants present. As this kit can be used to measure the total antioxidant capacity of plasma, serum, urine, saliva, or cell lysates, it is a great tool for evaluating the capacity to counter oxidative stress.

Combined Screening and End Product Assays

Several chemical compounds are produced during oxidative damage and may be detected and measured in many biological sites, providing different types of information. For example, they may be reliably measured, non-invasively, in urine, which screens for oxidative damage but provides little information regarding where or when the damage occurred. The same assay may also be used with samples at, or closer to, the site of production for that important information. These are some of Cayman's most popular assay kits.

8-isoprostane, produced by the random oxidation of tissue phospholipids, is a biomarker of lipid peroxidation. The **8-Isoprostane EIA Kit (Catalog No. 516351)** can be used to screen for changes in this biomarker in urine, serum, or plasma prior to more focused studies in tissues and cells. Importantly, this kit can be used appropriately in those focused studies to quantify tissue and cellular changes. Also, isoprostanes appear as artifacts in tissue and plasma samples which have undergone oxidative degradation during prolonged or improper storage, providing yet another application for this kit.

While 8-isoprostane is a well-established biomarker, the isoprostane $iPF_{2\alpha}$-VI is a more abundant product during oxidative stress and may, in fact, be a better biomarker. The **$iPF_{2\alpha}$-VI EIA Kit (Catalog No. 516301)** is the first simple, reliable EIA method for the measurement of this prominent product. It can be used for the quantification of $iPF_{2\alpha}$-VI from urine, plasma, cultured cells, and tissues. Like the 8-Isoprostane EIA Kit, this EIA can be used to screen for changes in the urine before progressing into more detailed studies within the body.

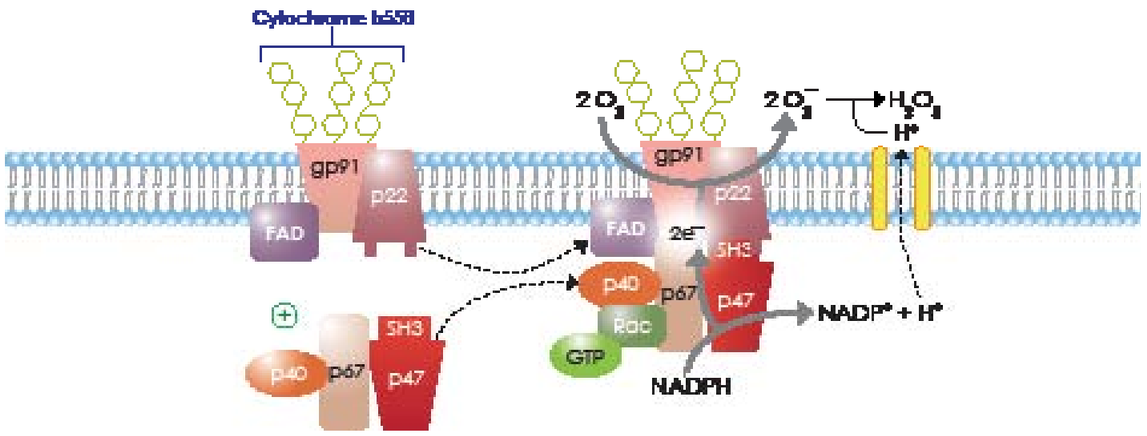


Figure 1. Reactive oxygen species generated by NADPH oxidase

Under conditions of oxidant stress such as that induced by either cigarette smoking or hypercholesterolemia, *ent*-$PGF_{2\alpha}$ increases disproportionally in relation to its enantiomer $PGF_{2\alpha}$. The ***ent*-Prostaglandin $F_{2\alpha}$ EIA Kit (Catalog No. 10010382)** is a competitive EIA that can be used for quantification of *ent*-$PGF_{2\alpha}$ in urine and other sample matrices.

Another established biomarker of oxidative stress is 8-hydroxy-2-deoxy guanosine (8-OH-dG), a product of oxidative damage of DNA by reactive oxygen and nitrogen species. The **8-hydroxy-2-deoxy Guanosine EIA Kit (Catalog No. 589320)** can be used to screen for changes in 8-OH-dG in urine samples, which occurs in response to xenobiotics as well as during diseases including cancer, diabetes, and hypertension. This competitive EIA can also be used for the quantification of 8-OH-dG in cell culture, plasma, and other sample matrices to further study oxidative damage to DNA.

The production of reactive nitrogen species can occur during oxidative stress. Changes in nitrite (NO_2^-) and nitrate (NO_3^-) can be detected in urine as well as in plasma/serum, tissue culture media, and tissue homogenates using the **Nitrate/Nitrite Colorimetric Assay Kit (Catalog No. 780001)**. This assay measures total nitrate/nitrite concentration using Griess reagents. Note that this assay cannot be used for *in vitro* assays of nitric oxide synthase (NOS) in which excess NADPH has been added, as NADPH interferes with the chemistry of the Griess reagents. For these assays a second method (LDH method) is utilized (Catalog No. 760871). These assays are ideal for the measurement of large nitrate/nitrite levels; the more sensitive fluorometric assay, described below, should be used for quantifying low levels.

Assays for End Products

Several chemical products and chemical modifications are indicative of the presence, absence, or severity of oxidative stress. The Cayman assay kits (both those directly below and those above) let you measure many of those chemical changes.

As mentioned above, H_2O_2 and reactive nitrogen species increase during oxidative stress. The detection of H_2O_2 at the cellular level pinpoints a source of production of this reactive oxygen species. The **Hydrogen Peroxide Cell-Based Assay Kit (Catalog No. 600050)** allows sensitive quantitation of H_2O_2 in cultured cells. The **Nitrate/Nitrite Fluorometric Assay Kit (Catalog No. 780051)** provides a convenient method for the quantitation of low levels of nitrite and nitrate in biological samples (particularly tissue culture medium). Nitrate/nitrite concentrations in a variety of samples can be accurately quantitated using this kit as it extends the lower limit of detection by 20-fold over our popular colorimetric version.

Glutathione (GSH), on the other hand, is a major cellular antioxidant that can be oxidized to the disulfide dimer GSSG. Normally, GSH is abundant as GSSG is rapidly reverted to GSH by glutathione reductase. Cayman's **Glutathione Assay Kit (Catalog No. 703002)** measures both GSH and GSSG, determining total glutathione to assess whether there is a deficiency in this peptide. The kit can also be used to measure only GSSG by following an alternative protocol. GSH measurement can be done in plasma, tissue samples, and cultured cells using this kit.

Protein modification occurs during oxidative stress. The formation of carbonyl groups (*e.g.*, aldehyde or ketone groups) to proteins is a validated biomarker of oxidative stress. The **Protein Carbonyl Assay Kit (Catalog No. 10005020)** is a popular assay kit for the measurement of this biomarker in plasma, serum, cell lysates, and tissue homogenates. S-Nitrosylation and S-glutathionylation refer to the binding of reactive nitrogen and oxidized glutathione, respectively, to thiol (-SH) side chains of protein cysteine residues. These reversible post-translational modifications increase with oxidative stress, regulating the activity of a large number of targets, including metabolic, structural, cytoskeletal, and signaling proteins. The **S-Nitrosylated Protein Detection Kit (Catalog No. 10006518)** and the **S-Glutathionylated Protein Detection Kit (Catalog No. 10010721)** provide reagents necessary for evaluating these changes.

Aldehydes increase dramatically on DNA as well as on proteins during oxidative stress. These can be detected using an aldehyde reactive probe (ARP), like the one used in Cayman's **Aldehyde Site (DNA and Protein) Detection Kit (Catalog No. 600170)**. This kit, designed for measuring total aldehydes in whole cells, can be adapted to analyze isolated DNA and protein samples.

Finally, lipid peroxidation results in the formation of highly reactive and unstable hydroperoxides of both saturated and unsaturated lipids. The **Lipid Hydroperoxide (LPO) Assay Kit (Catalog No. 705002)** measures hydroperoxides directly, utilizing the redox reactions with ferrous ions. An easy to use quantitative extraction methods eliminates any interference caused by H_2O_2 or endogenous ferric ions in the sample, providing a sensitive and reliable assay for lipid peroxidation.

Enzymes

Enzymes play pivotal roles in producing, preventing, and fixing damage produced by free radicals. These assays measure their activity.

One producing enzyme, xanthine oxidase, generates superoxide, a powerful reactive oxygen species, when oxidizing NADH. Levels of circulating xanthine oxidase can increase dramatically with disease. Cayman's **Xanthine Oxidase Assay Kit (Catalog No. 10010895)** provides a simple and accurate method for quantifying xanthine oxidase activity. Similarly, NOS generate nitric oxide, which can interact with superoxide to produce reactive nitrogen species. The **NOS Activity Assay Kit (Catalog No. 781001)** measures NOS activity by monitoring the conversion of radiolabeled arginine to citrulline. This assay is simple, sensitive, and specific for NOS activity and can be used with both crude and purified enzyme preparations.

Damage preventing enzymes include the superoxide dismutases (SOD), which convert superoxide into oxygen and H_2O_2, and catalase, which catalyzes the conversion of H_2O_2 to molecular oxygen and water. The **Superoxide Dismutase Assay Kit (Catalog No. 706002)** is a fast and reliable assay for the measurement of SOD activity from plasma, serum, tissue homogenates, and cell lysates. The **Catalase Assay Kit (Catalog No. 707002)** can be used to measure catalase activity in plasma, serum, erythrocyte lysates, tissue homogenates, and cell lysates. A third group of protective enzymes are the thioredoxin reductases (TrxR), which reduce thioredoxin, a small oxidoreductase enzyme containing a dithiol-disulfide active site to reduce numerous proteins. The **Thioredoxin Reductase Assay Kit (Catalog No. 10007892)** includes all reagents needed to assay mammalian TrxR activity.

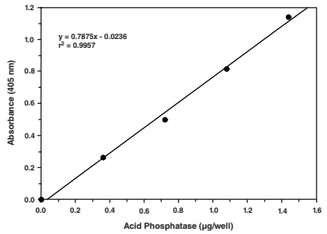
Several enzymes are involved in glutathione processing. As mentioned above, glutathione reductase catalyzes the reduction of oxidized glutathione (GSSG) to glutathione (GSH). The **Glutathione Reductase Assay Kit (Catalog No. 703202)** can be used to measure this important activity in cell lysates, tissue homogenates, and plasma. A family of glutathione peroxidases catalyzes the reduction of hydroperoxides, including H_2O_2, by reduced glutathione and functions to protect the cell from oxidative damage. The **Glutathione Peroxidase Assay Kit (Catalog No. 703102)** measures this activity indirectly by a coupled reaction with glutathione reductase. It can be used to measure all of the glutathione-dependent peroxidases in plasma, erythrocyte lysates, tissue homogenates, and cell lysates. Glutathione S-transferases protect cells against xenobiotics by conjugating reduced glutathione to electrophilic sites on the compound. The **Glutathione S-Transferase Assay Kit (Catalog No. 703302)** can be used to measure this activity in plasma, erythrocyte lysates, tissue homogenates, and cell lysates.

Miscellaneous

Acid Phosphatase Assay Kit 10008051

AP
Stability: ≥6 months at 4°C
Summary: Cayman's Acid Phosphatase Assay provides a method for detecting AP activity in plasma, serum, urine, and semen. The assay utilizes *para*-nitrophenyl phosphate (*p*NPP) as a chromogenic substrate for the enzyme. In the first step, AP dephosphorylates *p*NPP. In the second step, the phenolic OH-group is deprotonated under alkaline conditions resulting in *p*-nitrophenolate that yields an intense yellow color which can be measured at 405-414 nm.

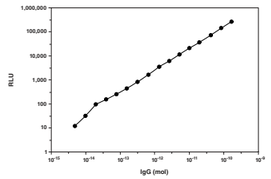
480 wells



Acridinium Protein Labeling Kit 200201

Stability: ≥1 year at 4°C
Summary: The Acridinium Protein Labeling contains all the reagents and instructions required to produce chemiluminescent-labeled proteins following a simple one-step procedure. The acridinium compound supplied in the kit contains an NHS ester which will react with primary amines to form a stable amide bond. Solutions for the detection of the labeled protein are also included.

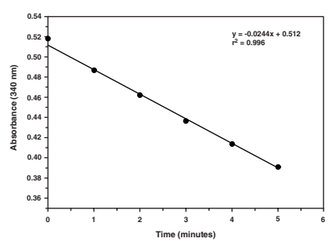
1 ea



Alanine Transaminase Activity Assay Kit 700260

ALT, Alanine Aminotransferase, ALAT, Serum Pyruvic Transaminase, sGPT
Stability: ≥6 months at -80°C
Summary: ALT is a homodimeric cytoplasmic pyridoxal phosphate-dependent enzyme involved in cellular nitrogen metabolism, amino acid metabolism, and liver gluconeogenesis. Cayman's Alanine Transaminase Assay provides a method of detecting ALT activity in serum, plasma, tissue samples, and cell lysates. Measurement of the ALT activity is carried out by monitoring the rate of NADH oxidation in a coupled reaction system employing LDH.

96 wells



ASP EIA Kit† 10008673

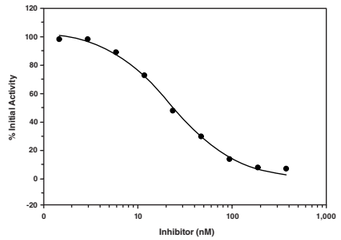
A31300401, Amnesic Shellfish Poison
Stability: ≥3 months at 4°C
Sensitivity: 80% B/B_0: 10 pg/ml
Summary: Domoic acid (DA) and DA derivatives are water-soluble neurotoxins produced by a number of marine algae, in particular those of the genus *Pseudo-nitzschia*. This ASP ELISA is specific for DA with no cross-reactivity to non-toxic, structural analogues like kainic acid, L-glutamic acid, L-glutamine, formimino-L-glutamic acid, proline, or γ-aminobutyric acid (GABA). The assay is primarily intended for use in routine monitoring of DA levels in bivalve molluscs to comply with the regulatory maximum permitted level, but is also applicable for DA quantification in other marine matrices.

96 wells

BACE Inhibitor Screening Assay Kit 600070

β-Secretase
Stability: ≥6 months at -80°C
Summary: BACE is a promising therapeutic target as this protease initiates the first step in Aβ production. Cayman's BACE Inhibitor Screening Assay provides a method for screening human BACE inhibitors. The assay utilizes a synthetic Swedish mutant APP peptide (EVNLDAEF) that has been linked to a fluorophore (EDANS) at one end and to a quenching agent (Dabcyl) at the other. After cleavage by BACE, the product (peptide-EDANS) is brightly fluorescent.

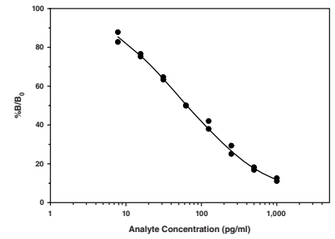
96 wells



Cayman Practice EIA Kit 10009658

Stability: ≥1 year at -20°C
Summary: This assay has been developed for researchers that do not have experience performing EIAs. It can be used as a practice tool allowing you to become comfortable with running Cayman EIAs. This kit contains enough reagents to run at least four complete standard curves.

96 wells

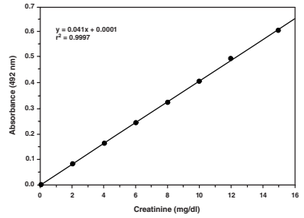


Biosense products are available through Cayman Chemical only within North & South America and Asia; elsewhere contact Biosense.

Creatinine Assay Kit 500701

Stability: ≥1 year at 4°C
Summary: Urinary creatinine levels are commonly used as an index of standardization for a variety of other tests. Measurement of creatinine clearance is also useful in detecting renal disease and estimating the extent of impairment of renal function. Cayman's Creatinine Assay features a 96-well plate format and has been validated for urine samples.

96 wells
480 wells



DNA Laddering Kit 660990

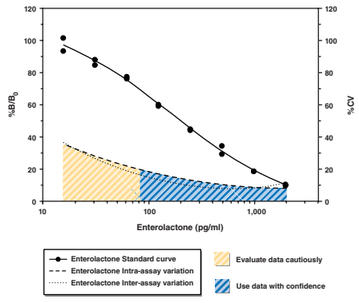
Stability: ≥1 year at -20°C
Summary: Apoptosis is associated with the fragmentation of chromosomal DNA into multiples of the 180 bp nucleosomal unit, known as DNA laddering. This kit is designed to maximize extraction of small fragments of DNA with minimal contamination from intact chromatin. The isolated DNA is separated by electrophoresis and can be visualized using ethidium bromide.

24 reactions

Enterolactone EIA Kit 500520

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 240 pg/ml • 80% B/B_0: 70 pg/ml
Summary: Enterolactone is a mammalian lignan with an estrogen-like diphenolic structure. It is produced by intestinal bacteria from two plant precursors (matairesinol and secoisolariciresinol) obtained in the diet. Enterolactone and other lignans and phytoestrogens have been associated with a reduced risk of acute coronary events, hormone-dependent cancers, and possibly osteoporosis. Several studies have suggested that serum enterolactone may serve as a biomarker of a healthy, high-fiber diet.

96 wells
480 wells



•Also Available: **Enterolactone EIA Kit (Solid Plate)** (10006647)

SPI-BIO products are available through Cayman Chemical only within North & South America and Asia; elsewhere contact SPI-BIO.

Formaldehyde Assay Kit 700380

Stability: ≥6 months at -20°C
Summary: Cayman's Formaldehyde Assay provides a fluorescence-based method for detecting formaldehyde in urine. The cyclization between formaldehyde and acetoacetanilide in the presence of ammonia results in a fluorescent product which is analyzed using an excitation wavelength between 365-375 nm and an emission wavelength between 465-475 nm.

96 wells

GFAP (human) EIA Kit+ 10007621

Glial Fibrillary Acidic Protein
Stability: ≥6 months at 4°C **Limit of Detection:** 0.04 ng/ml
Summary: GFAP is a 40-53 kDa monomeric molecule found only in adult glial cells of the central nervous system (CNS) and represents the major part of the cytoskeleton of astrocytes. GFAP is considered to be a reliable cell-specific biomarker for monitoring neuronal activity under developmental and pathological conditions, such as brain injury, and retinal stress. The GFAP (human) EIA can be used to measure GFAP in human samples such as serum, plasma, culture supernatant, and cerebrospinal fluid.

96 wells

His-Express Detection EIA Kit 10012445

Stability: ≥6 months at -20°C **Sensitivity:** ~100 ng/ml
Summary: Cayman's His-Express Detection EIA is competitive assay designed for the rapid, semi-quantitative screening of cell lysates and affinity column fractions for His-tagged proteins. It is intended to serve as a substitute for SDS-PAGE, thereby expediting the screening of affinity column fractions.

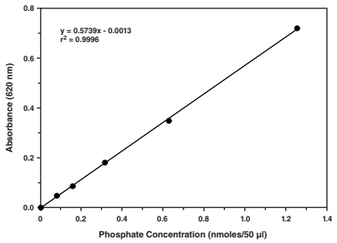
96 wells
480 wells

•Also Available: **His-Express Detection EIA Kit (Solid Plate)** (500002)

Malachite Green Phosphate Assay Kit 10009325

Stability: ≥6 months at 4°C
Summary: Cayman's Malachite Green Phosphate Assay provides a fast and reproducible, colorimetric method for measuring inorganic free phosphate in aqueous solutions. Applications for this assay include quantification of phosphorylation and phosphate release from protein phosphatase substrates. The assay is formatted to a 96-well format, but could easily be modified for use in 384-well or cuvette-based assays.

96 wells



Nuclear Extraction Kit 10009277

Please see the **Transcription Factor Assays** section for full listing on page 58

P-Glycoprotein Drug Interaction Assay Kit+ 789201

P-gp
Stability: ≥1 year at -20°C
Summary: P-gp is an active plasma membrane transporter involved in drug pharmacokinetics and cellular detoxification. This assay provides an *in vitro* screening method for testing drug interaction with P-gp based on the study of modulation of basal or induced ATPase activity from enriched P-gp membrane vesicle preparations.

96 wells

PBMC Fluorescent Titer Assay Kit+ 10004916

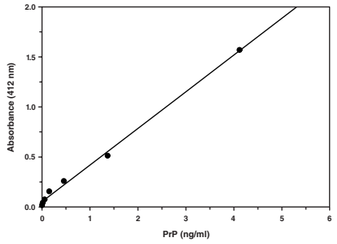
Peripheral Blood Mononuclear Cells
Stability: ≥6 months at -20°C
Summary: The PBMC Fluorescent Titer Assay is a patented method for quantifying the number of PBMCs based on fluorescent detection. The assay procedure involves cell lysis and PBMC quantification in the pellet using an intercalating substrate followed by fluorescence detection.

192 wells

Prion Protein EIA Kit+ 589751

Stability: ≥6 months at -20°C
Summary: This EIA is based on a double-antibody sandwich technique and has been validated for the detection of native cellular prion protein (PrP^c) in brain extracts. It can also be used to detect PrP^c extracted from other tissues, as well as denaturated PrP and recombinant PrP. The antibodies used in this kit were raised against SAF from hamster brain and crossreact with PrP from most mammalian species including murine, human, ovine, and cattle.

96 wells



Protein Determination Kit 704002

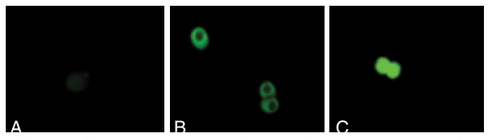
Stability: ≥1 year at 4°C **Limit of Detection:** 5.6 µg/ml
Summary: Cayman's Protein Determination is a microplate-based, colorimetric method for rapid total protein quantification.

480 wells

S-Glutathionylated Protein Detection Kit 10010721

PSSG
Stability: ≥1 year at -20°C
Summary: Cayman's PSSG Kit provides a method for the direct visualization of S-glutathionylated proteins in whole (permeabilized) cells by flow cytometry and microscopy as well as avidin overlay analysis. This assay starts with the modification of protein free-thiols groups followed by enzymatic cleavage of any PSSG adducts present in the sample. Biotinylation of the newly-formed protein free-thiols provides the basis for visualization using streptavidin-based colorimetric or fluorescence detection.

1 ea



Typical immunfluorescence images using murine monocytes. *Panel A:* Cells stained by the standard method with omission of Reduction Reagent generated no fluorescence. *Panel B:* Cells stained by the method as written reveal S-glutathionylated proteins. *Panel C:* Cells treated by the method with omission of free-thiol Blocking Reagent reveals labeling of all accessible protein thiols.

+ SPI-BIO products are available through Cayman Chemical only within North & South America and Asia; elsewhere contact SPI-BIO.

SureLight® Western Kits

Save time and money with Columbia Bioscience's Western Blot Detection kits. SureLight® Western kits provide the same sensitivity as enzymatic chemiluminescence (ECL) and enzymatic chemifluorescence (ECF) reagents. These assays are up to 16X more sensitive than other fluorescent kits. So, you can lose the enzyme and save up to an hour in processing your blots but keep the sensitivity. Sufficient buffers and detection reagent are provided for up to ten 10 x 10 cm^2 blots.

SureLight® Western Kit (P112) 13749

Stability: ≥6 months at 4°C
Summary: The Western Kit (P112) features a goat anti-mouse SureLight® P3 conjugate.

1 ea

SureLight® Western Kit (R112) 13768

Stability: ≥6 months at 4°C
Summary: The Western Kit (R112) features a goat anti-mouse R-Phycoerythrin conjugate.

1 ea

SureLight® Western Kit (P114) 13767

Stability: ≥6 months at 4°C
Summary: The Western Kit (P114) features a goat anti-rabbit SureLight® P3 conjugate.

1 ea

SureLight® Western Kit (R114) 13769

Stability: ≥6 months at 4°C
Summary: The Western Kit (R114) features a goat anti-rabbit R-Phycoerythrin conjugate.

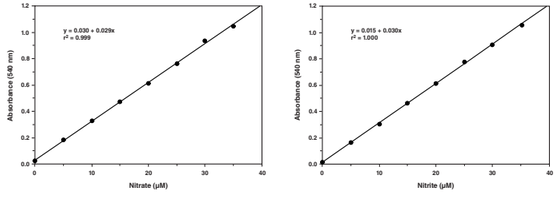
1 ea

Nitric Oxide

Nitrate/Nitrite Colorimetric Assay Kit 780001

Nitric Oxide Metabolite Detection Kit
Stability: ≥1 year at -20°C
Summary: Cayman's Nitrate/Nitrite Assay provides an accurate and convenient method for measurement of total nitrate/nitrite concentrations. This kit can be used to measure nitrate and nitrite in plasma/serum, urine, tissue culture media, and tissue homogenates.

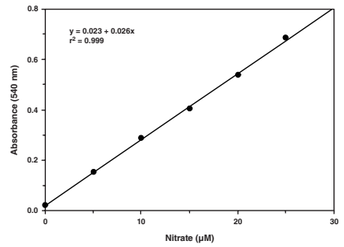
192 wells



Nitrate/Nitrite Colorimetric Assay Kit (LDH method) 760871

Nitric Oxide Metabolite Detection Kit
Stability: ≥1 year at -20°C
Summary: NADPH is an essential cofactor for the function of the NOS enzyme. Unfortunately, NADPH interferes with the chemistry of the Griess reagents, which are the most commonly used reagents for nitrite detection. This kit uses Lactate Dehydrogenase (LDH) to oxidize the excess NADPH used in a NOS-catalyzed reaction, thereby making the assay particularly well suited to measurements of NOS activity *in vitro*.

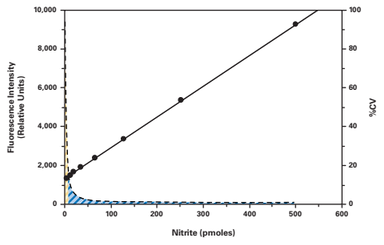
96 wells



Nitrate/Nitrite Fluorometric Assay Kit 780051

Nitric Oxide Metabolite Detection Kit
Stability: ≥1 year at -20°C
Summary: Cayman's Nitrate/Nitrite Fluorometric Assay provides a method for the quantitation of low levels of nitrate and nitrite in biological samples (particularly tissue culture medium). The minimum detectable quantity of NO_2^-/NO_3^- is ~50 nM.

2 x 96 wells



S-Nitrosylated Protein Detection Kit 10006518

SNO
Stability: ≥1 year at -20°C
Summary: Cayman's S-Nitrosylated Protein Detection Assay employs a modification of the Jaffrey, *et al.* 'Biotin-switch' method to allow for the direct visualization of S-nitrosylated proteins in whole cells or tissues, as well as by western blot analysis. Using this method, free -SH groups are first blocked and any S-NO bonds present in the sample are then cleaved. Biotinylation of the newly formed free -SH groups provides the basis for visualization using streptavidin-based colorimetric or fluorescence detection.

1 ea

NOS Activity Assay Kit 781001

Stability: ≥1 year at -80°C
Summary: The NOS Activity Assay measures NOS activity by monitoring the conversion of radiolabeled arginine to citrulline. This assay is simple, sensitive, and specific for NOS activity and can be used with both crude and purified enzyme preparations. The kit includes sufficient materials and reagents for 50 total reactions. Radiolabeled arginine and NADPH are not included with the kit.

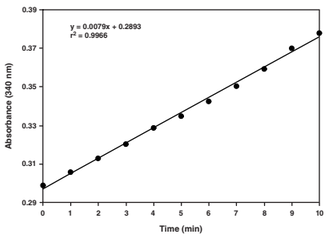
1 ea

Oxidative Injury

Aconitase Assay Kit 705502

Stability: ≥6 months at -20°C
Summary: Aconitase is an iron-sulfur protein containing a $[Fe_4S_4]^{2+}$ cluster that catalyzes the stereospecific isomerization of citrate to isocitrate *via cis*-aconitate. Whereas exposure of aconitase to oxidants renders the enzyme inactive, loss of aconitase activity in cells or in biological samples treated with pro-oxidants has been interpreted as a measure of oxidative damage. Cayman's Aconitase Assay provides a simple, reproducible, and sensitive tool for assaying aconitase from tissue homogenates or cell lysates.

96 wells



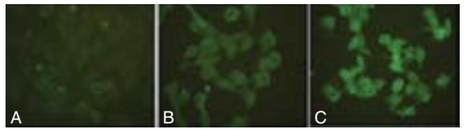
Recommendations for Nitrate/Nitrite Assay Kits

Sample Type	Nitrate/Nitrite Colorimetric Assay Kit	Nitrate/Nitrite Flurometric Assay Kit	Nitrate/Nitrite Colorimetric Assay Kit (LDH Method)
Plasma/Serum	✓	sample purification required	not recommended
Cell Culture - iNOS (High [NOx] > 2 µM	✓	not recommended	not recommended
Cell Culture - eNOS (Low [NOx] < 2 µM	not suitable	✓	not suitable
***In Vitro* NOS Assays**	not suitable	not suitable	✓

Aldehyde Site (DNA and Protein) Detection Kit 600170

Stability: ≥1 year at -20°C
Summary: Oxidative stress in cells can result in the appearance and accumulation of free aldehydes on DNA and proteins. Cayman's Aldehyde Site (DNA and Protein) DetectionKit employs an aldehyde reactive probe, O-(biotinylcarbazoylmethyl) hydroxylamine, that reacts specifically with aldehyde groups resulting from protein or DNA modification. Modified sites are then detected using avidin-conjugated reporters.

96 wells

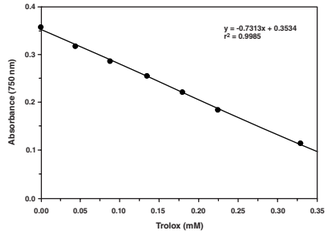


Hydrogen peroxide induces oxidative damage in HeLa cells. HeLa cells were treated with vehicle *(Panel A)*, 0.015% (4.9 mM) H_2O_2 *(Panel B)*, and 0.03% (9.8 mM) H_2O_2 *(Panel C)* for four hours and then processed for staining.

Antioxidant Assay Kit 709001

Stability: ≥1 year at 4°C
Summary: Cayman's Antioxidant Assay measures the total antioxidant capacity of plasma, serum, urine, saliva, or cell lysates. The assay relies on the ability of antioxidants in the sample to inhibit the oxidation of ABTS to $ABTS^{\bullet+}$ by metmyoglobin. The capacity of the antioxidants in the sample to prevent ABTS oxidation is compared with that of Trolox, a water-soluble tocopherol analog, and is quantified as molar Trolox equivalents.

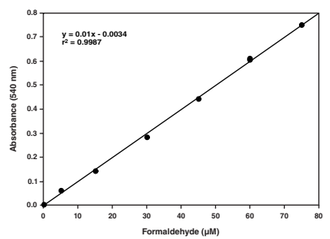
96 wells



Catalase Assay Kit 707002

CAT
Stability: ≥1 year at 4°C
Summary: Catalase is an ubiquitous antioxidant enzyme that is responsible for the detoxification of H_2O_2. Cayman's Catalase Assay utilizes the peroxidatic function of catalase for determination of enzyme activity. The assay can be used to measure catalase activity in plasma, serum, erythrocyte lysates, tissue homogenates, and cell lysates.

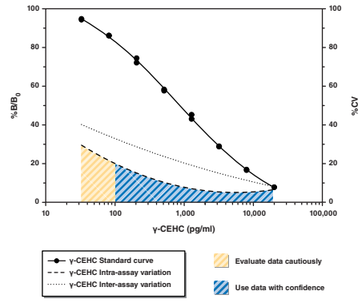
96 wells
480 wells



γ-CEHC EIA Kit (plasma and serum) 10010621

2,7,8-trimethyl-2-(β-Carboxy-Ethyl)-6-Hydroxychroman
Stability: ≥6 months at -20°C
Summary: γ-Tocopherol is the most abundant form of vitamin E in the diet. Its metabolite, γ-CEHC, is produced in the liver by the action of CYP450 enzymes and excreted in urine at levels that exceed all other tocopherol metabolites. Cayman's γ-CEHC EIA can be used for efficient quantification of γ-CEHC in plasma and serum.

96 wells
480 wells

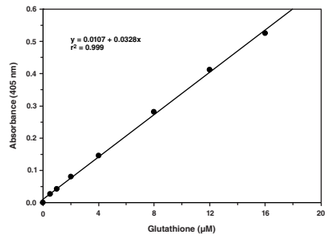


• Also Available: **γ-CEHC EIA Kit (plasma and serum) (Solid Plate)** (10011717)

Glutathione Assay Kit 703002

GSH
Stability: ≥1 year at 4°C
Summary: Cayman's GSH Assay utilizes a carefully optimized enzymatic recycling method for the quantification of GSH in a 96-well microplate format. It measures both GSH and GSSG to reflect total glutathione in a sample. The kit can also be used to measure only GSSG by following an alternative protocol. The GSH Assay can be used for plasma, tissue samples, and cultured cells with minimal sample processing.

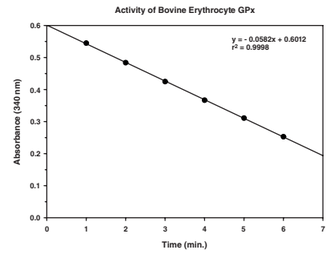
96 wells
480 wells



Glutathione Peroxidase Assay Kit 703102

GPx
Stability: ≥6 months at -20°C
Summary: GPx catalyzes the reduction of hydroperoxides, including H_2O_2, using reduced glutathione and thereby functions to protect the cell from oxidative damage. Cayman's Glutathione Peroxidase Assay measures GPx activity indirectly by a coupled reaction with glutathione reductase (GR). Cayman's GPx Assay can be used to measure all of the glutathione-dependent peroxidases in plasma, erythrocyte lysates, tissue homogenates, and cell lysates.

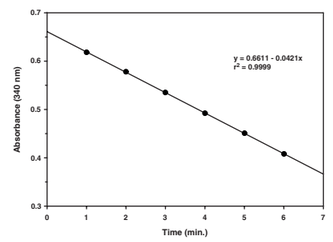
96 wells
480 wells



Glutathione Reductase Assay Kit 703202

GR
Stability: ≥6 months at -20°C
Summary: GR is a flavoprotein that catalyzes the NADPH-dependent reduction of oxidized GSSG to GSH. This enzyme is essential for the GSH redox cycle which maintains adequate levels of reduced cellular GSH, which is essential for protection against oxidative stress. Cayman's Glutathione Reductase Assay Kit measures GR activity by measuring the rate of NADPH oxidation.

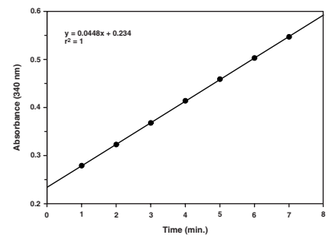
96 wells



Glutathione S-Transferase Assay Kit 703302

GST
Stability: ≥1 year at -20°C
Summary: GSTs are ubiquitous multifunctional enzymes, which play a key role in cellular detoxification. Cayman's GST Assay measures total GST activity (cytosolic and microsomal) by measuring the conjugation of 1-chloro-2,4-dinitrobenzene (CDNB) with reduced glutathione. Cayman's GST Assay can be used to measure GST activity in plasma, erythrocyte lysates, tissue homogenates, and cell lysates. Cytosolic and microsomal GST activity can also be individually assayed.

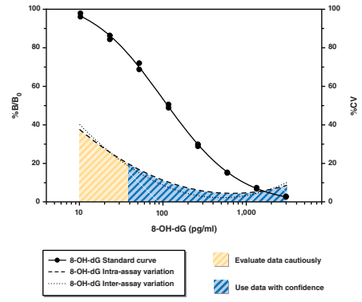
96 wells



8-hydroxy-2-deoxy Guanosine EIA Kit 589320

8-OH-dG
Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 100 pg/ml • 80% B/B_0: 30 pg/ml
Summary: 8-OH-dG is a product of oxidative damage of DNA by reactive oxygen and nitrogen species and serves as an established marker of oxidative stress. Cayman's 8-OH-dG EIA is a competitive assay that can be used for the quantification of 8-OH-dG in urine, cell culture, plasma, and other sample matrices. The EIA utilizes an anti-mouse IgG-coated plate and a tracer consisting of an 8-OH-dG AChE conjugate. This format has the advantage of providing low variability and increased sensitivity compared to assays that utilize an antigen-coated plate.

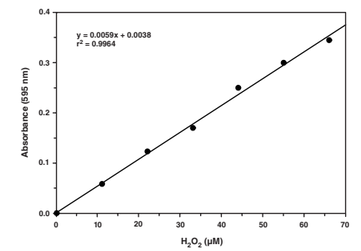
96 wells
480 wells



Hydrogen Peroxide (urinary) Assay Kit 706011

H_2O_2
Stability: ≥1 year at 4°C
Summary: H_2O_2 is a ubiquitous, toxic, metabolic by-product of aerobic respiration. Cayman's H_2O_2 Assay utilizes the well-established xylenol orange detection method for quantifying the oxidation of ferrous ions (Fe^{2+}) to ferric ions (Fe^{3+}) by H_2O_2. A unique feature of Cayman's assay is the inclusion of catalase as an H_2O_2 scavenger for the purpose of confirming the specificity of the reaction for H_2O_2. The sensitivity and the specificity of the assay make it well-suited to accurately measure urinary levels of H_2O_2.

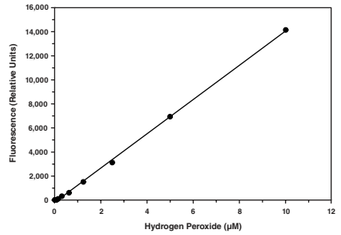
96 wells



Hydrogen Peroxide Cell-Based Assay Kit 600050

Stability: ≥6 months at -20°C
Summary: It is well established that H_2O_2 is a cytotoxic agent but evidence also suggests that H_2O_2 may be an important regulator of eukaryotic signal transduction. Cayman's H_2O_2 Cell-Based Assay provides a simple fluorometric method for the sensitive quantitation of H_2O_2 in cultured cells. H_2O_2 is detected using ADHP in the presence of horseradish peroxidase to produce highly fluorescent resorufin. Catalase, an H_2O_2 scavenger, is included in the kit for checking specificity of the assay.

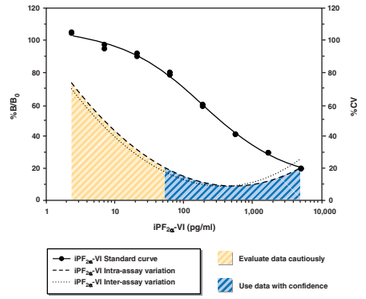
96 wells
480 wells



$iPF_{2\alpha}$-VI EIA Kit 516301

Stability: ≥6 months at -20°C
Sensitivity: 50% B/B_0: 250 pg/ml • 80% B/B_0: 50 pg/ml
Summary: This assay is the first EIA method for the measurement of the more abundant $iPF_{2\alpha}$-VI isoprostane. Normal urinary levels of $iPF_{2\alpha}$-VI are 500-700 pg/mg creatinine. Cayman's $iPF_{2\alpha}$-VI EIA is a competitive assay that can be used for the quantification of $iPF_{2\alpha}$-VI from plasma, urine, cultured cells, and tissues.

96 wells
480 wells



•Also Available: **$iPF_{2\alpha}$-VI EIA Kit (Solid Plate)** (516301.1)

8-Isoprostane Affinity Purification Kit (4 ml) 10367

$iPF_{2\alpha}$-III, 8-epi $PGF_{2\alpha}$, 8-iso $PGF_{2\alpha}$
Stability: ≥2 years at 4°C
Summary: This kit contains all reagents necessary for simple one-step purification of 8-isoprostane from most biological samples.

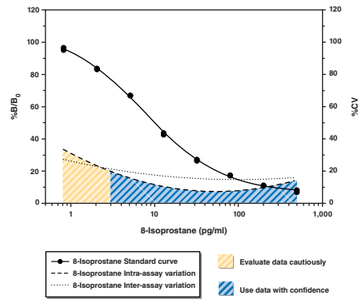
1 ea
5 ea

•Also Available: **8-Isoprostane Affinity Column** (10010366)
8-Isoprostane Affinity Purification Kit (20 ml) (10368)
8-Isoprostane Affinity Sorbent (10010365)

8-Isoprostane EIA Kit 516351

$iPF_{2\alpha}$-III, 8-epi $PGF_{2\alpha}$, 8-iso $PGF_{2\alpha}$
Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 10 pg/ml • 80% B/B_0: 2.7 pg/ml
Summary: The isoprostanes are a family of eicosanoids of non-enzymatic origin produced by the random oxidation of tissue phospholipids by oxygen radicals. 8-Isoprostane has been proposed to be a marker of antioxidant deficiency and oxidative stress. Plasma from healthy volunteers contains modest amounts of 8-isoprostane (40-100 pg/ml) that increase with the age of the test subject. Normal human urinary levels range from 10-50 ng/mmol creatinine.

96 wells
480 wells

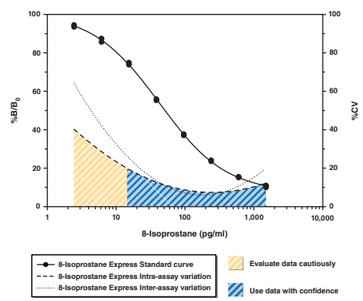


•Also Available: **8-Isoprostane EIA Kit (Solid Plate)** (516351.1)

8-Isoprostane Express EIA Kit 516360

$iPF_{2\alpha}$-III, 8-epi $PGF_{2\alpha}$, 8-iso $PGF_{2\alpha}$
Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 50 pg/ml • 80% B/B_0: 10 pg/ml
Summary: This assay offers the convenience of a fast assay (2 hour incubation; 1 hour development) while still achieving a detection limit (80% B/B_0) of 10 pg/ml.

96 wells
480 wells

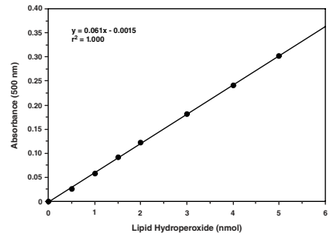


•Also Available: **8-Isoprostane Express EIA Kit (Solid Plate)** (516361)
STAT-8-Isoprostane EIA Kit (500431)

Lipid Hydroperoxide (LPO) Assay Kit 705002

Stability: ≥1 year at 4°C
Summary: Lipid peroxidation results in the formation of highly reactive, unstable hydroperoxides of both saturated and unsaturated lipids. Cayman's Lipid Hydroperoxide Assay measures lipid hydroperoxides utilizing the redox reaction with ferrous ions. An easy to use quantitative extraction method is used to extract lipid hydroperoxides into chloroform and then the extract is used directly in the assay. This kit is designed for use with either a single-tube spectrophotometer or with a 96-well microplate reader. The microplate reader requires a reusable glass plate that is supplied with the LPO Assay Kit (96 well) (Catalog No. 705003).

100 dtn



•Also Available: **Lipid Hydroperoxide (LPO) Assay Kit (96 well)** (705003)

Methionine Sulfoxide Immunoblotting Kit 600160

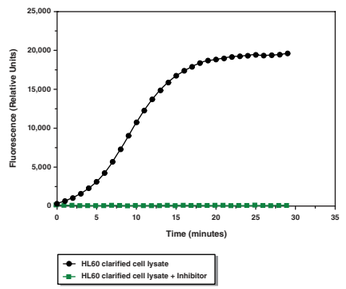
MetO
Stability: ≥1 year at -20°C
Summary: Protein MetO is a reversible oxidative modification that occurs by exposure of protein(s) methionine residues to reactive oxygen species (ROS). Cayman's MetO Immunoblotting Kit contains reagents needed for the immunochemical detection of proteins containing MetO residues by western blotting. MetO-containing samples of interest include those from cell or tissue lysates as well as semi-pure or purified proteins.

10 blots

Myeloperoxidase Chlorination Assay Kit 10006438

Stability: ≥6 months at -20°C
Summary: Cayman's Myeloperoxidase Chlorination Assay provides a fluorescence-based method for detecting the MPO chlorination activity in both crude cell lysates and purified enzyme preparations. The assay utilizes the non-fluorescent probe, APF, which is selectively cleaved by hypochlorite to yield the highly fluorescent compound fluorescein. The kit includes an MPO-specific inhibitor for distinguishing MPO activity from MPO-independent fluroescence.

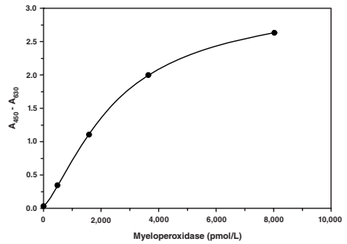
2 x 96 wells



Myeloperoxidase (human) EIA Kit 585001

Stability: ≥6 months at 4°C **Limit of Detection:** 14 pmol/L
Summary: Cayman's MPO (human) EIA is an immunometric assay which can be used to measure MPO in plasma without prior sample purification. This assay has been tested using plasma from healthy volunteers and the results were shown to be consistent with published data.

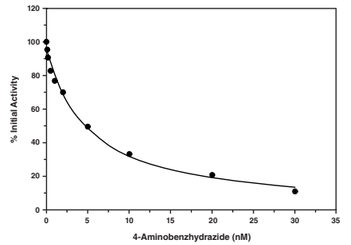
96 wells



Myeloperoxidase Inhibitor Screening Assay Kit 700170

Stability: ≥6 months at -20°C
Summary: Cayman's MPO Inhibitor Screening Assay provides fluorescence-based methods for screening inhibitors to both the chlorination and peroxidation activities of MPO. Sufficient reagents are provided for a full 96-well plate assay of each type of activity.

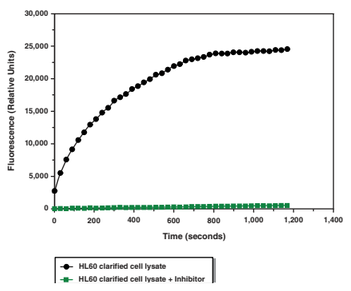
2 x 96 wells



Myeloperoxidase Peroxidation Assay Kit 700160

Stability: ≥6 months at -20°C
Summary: Cayman's MPO Peroxidation Assay provides a fluorescence-based method for detecting MPO peroxidase activity in both crude cell lysates and purified enzyme preparations. The MPO-catalyzed reaction between H_2O_2 and ADHP produces the highly fluorescent compound resorufin. The kit includes an MPO-specific inhibitor for distinguishing MPO activity from MPO-independent fluroescence.

2 x 96 wells



S-Nitrosylated Protein Detection Kit 10006518

Please see the **Nitric Oxide** section for full listing on page 37

oxLDL-β_2GPI (human) ELISA Kit 10007893

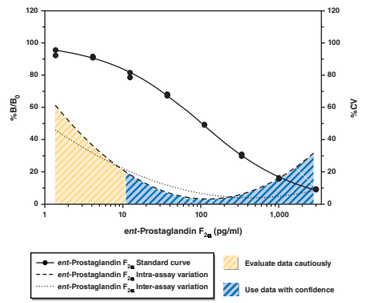
Oxidized LDL-β_2 Glycoprotein I (human)
Stability: ≥6 months at 4°C
Summary: oxLDL is the principal form of cholesterol that accumulates in atherosclerotic lesions or plaques. Unlike native LDL, oxLDL binds to β_2GPI to form oxLDL-β_2GPI complexes. Stable oxLDL-β_2GPI complexes are regarded as pathogenic and appear to be highly clinically relevant. Cayman's oxLDL-β_2GPI (human) ELISA is an immunometric (*i.e.*, sandwich) assay that detects the circulating oxLDL-β_2GPI complex in human serum or plasma.

1 ea

ent-Prostaglandin $F_{2\alpha}$ EIA Kit 10010382

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 110 pg/ml • 80% B/B_0: 20 pg/ml
Summary: The majority of $PGF_{2\alpha}$ found in urine is formed non-enzymatically, as its formation cannot be blocked by inhibitors of COX activity. Chiral LC and GC-MS demonstrate that much of the urinary $PGF_{2\alpha}$ is the enantiomer of $PGF_{2\alpha}$, *ent*-$PGF_{2\alpha}$. Under conditions of oxidant stress, ent-$PGF_{2\alpha}$ increases disproportionally in relation to $PGF_{2\alpha}$. Cayman's *ent*-$PGF_{2\alpha}$ Assay is a competitive EIA that can be used for quantification of *ent*-$PGF_{2\alpha}$ in urine and other sample matrices.

96 wells
480 wells



Protein Carbonyl Assay Kit 10005020

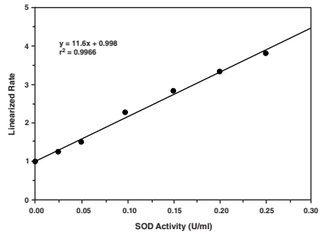
Stability: ≥1 year at 4°C
Summary: Cayman's Protein Carbonyl Assay is a colorimetric assay for the measurement of oxidized proteins. The carbonyls of protein samples are derivatized using 2,4-dinitrophenylhydrazine (DNPH). Formation of a Schiff base produces the corresponding hydrazone which can be analyzed spectrophotometrically at 360-385 nm. This assay can be used to measure oxidized proteins in plasma, serum, cell lysates, and tissue homogenates.

96 wells

Superoxide Dismutase Assay Kit 706002

SOD
Stability: ≥1 year at -20°C
Summary: Cayman's SOD Assay is a fast and reliable assay for the measurement of SOD activity from plasma, serum, tissue homogenates, and cell lysates. SOD activity is assessed by measuring the dismutation of superoxide radicals generated by xanthine oxidase and hypoxanthine in a convenient 96-well format. A key feature of the kit is the inclusion of a quality-controlled SOD standard. The standard curve generated using this enzyme provides a means to accurately quantify the activity of all three types of SOD (Cu/Zn-, Mn-, and Fe-SOD).

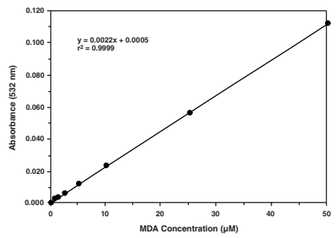
96 wells
480 wells



TBARS Assay Kit 10009055

Thiobarbituric Acid Reactive Substances
Stability: ≥1 year at 4°C
Summary: Decomposition of the unstable peroxides derived from oxidation of polyunsaturated fatty acids results in the formation of malondialdehyde (MDA), which can be quantified colorimetrically following its controlled reaction with thiobarbituric acid. The measurement of these TBARS is a well-established method for screening and monitoring lipid peroxidation. Cayman's TBARS Assay provides a simple, reproducible, and standardized tool for assaying lipid peroxidation in plasma, serum, urine, tissue homogenates, and cell lysates.

96 wells



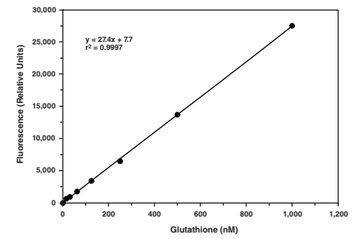
Recommendations for PGE_2 Assay Kits

Sample Type	**PGE_2 EIA Kit -Monoclonal** most sensitive; overnight incubation; most widely used assay	**PGE_2 EIA Express Kit** one hour incubation; 3 fold less sensitive than PGE_2 EIA Kit-Monoclonal	**PGEM EIA Kit** measures PGE_2 metabolites	**PGE_2 FPIA Kit Green or Red** homogenous, 1 step assay for HTS labs; least sensitive
Plasma/Serum	✓	not recommended	✓	not suitable
Urine	urinary PGE_2 produced directly by the kidney	not recommended	✓	not suitable
Tissues	✓	not recommended	✓	not suitable
Cell Culture	✓	✓	not applicable	✓
***In Vitro* Expression Systems**	✓	✓	not applicable	✓

Thiol Detection Assay Kit 700340

Stability: ≥6 months at -20°C
Summary: The detection and measurement of free thiols (*i.e.,* free cysteine, glutathione, and cysteine residues on proteins) is one of the essential tasks for investigating biological processes and events in many biological systems. Cayman's Thiol Detection Assay provides a simple, sensitive fluorometric method for assaying free thiol content in samples (*i.e.,* plasma, serum, tissue homogenates, cell lysates, and urine), using a proprietary fluorometric detector.

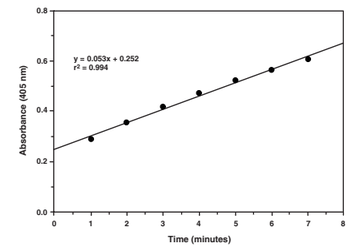
96 wells
480 wells



Thioredoxin Reductase Assay Kit 10007892

TrxR
Stability: ≥6 months at -20°C
Summary: Cayman's TrxR Assay provides a method for quantifying mammalian TrxR activity from tissue homogenates and cell lysates in a colorimetric 96-well plate format. In this assay, TrxR uses NADPH to reduce DTNB to 5-thio-2-nitrobenzoic acid (TNB) which absorbs strongly at 405-414 nm. Measurement of TrxR activity in the absence and in the presence of aurothiomalate, a specific TrxR inhibitor included in the kit, allows for correction of non-thioredoxin reductase-independent DTNB reduction.

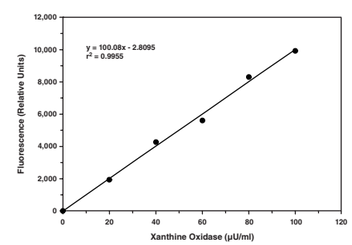
96 wells



Xanthine Oxidase Assay Kit 10010895

Xanthine Oxidoreductase, XO
Stability: ≥6 months at -20°C
Summary: XO catalyzes the hydroxylation of hypoxanthine to xanthine and then further catalyzes the oxidation of xanthine to uric acid. When oxidizing NADH, XO generates superoxide, a powerful ROS. Cayman's XO Assay provides a simple and accurate method for quantifying xanthine oxidase activity. The assay is based on a multistep enzymatic reaction resulting in generation of the highly fluorescent product resorufin.

96 wells

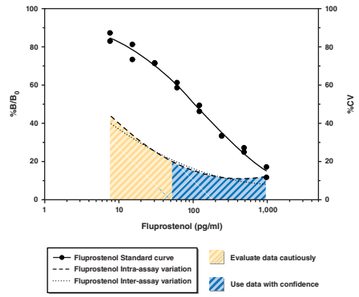


Prostaglandins

Fluprostenol EIA Kit 516761

16-m-trifluoromethylphenoxy tetranor $PGF_{2\alpha}$
Stability: ≥6 months at -20°C
Sensitivity: 50% B/B_0: 110 pg/ml • 80% B/B_0: 16 pg/ml
Summary: Fluprostenol is a metabolically stable analog of $PGF_{2\alpha}$ with potent FP receptor agonist activity. The isopropyl ester of fluprostenol has been approved for use as an ocular hypotensive drug for the treatment of glaucoma and is sold under the Alcon trade name Travoprost. Cayman's Fluprostenol EIA is a sensitive detection method for measuring the free acid form of fluprostenol. The assay also detects the isopropyl ester form of fluprostenol, but with less sensitivity. The assay has been validated for use in aqueous humor samples.

96 wells
480 wells



• Also Available: **Fluprostenol EIA Kit (Solid Plate)** (516761.1)

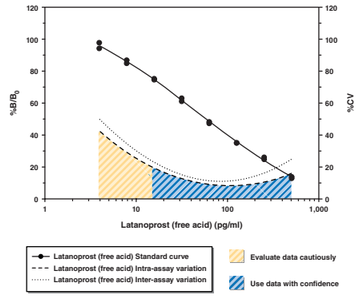
8-Isoprostane EIA Kit 516351

Please see the **Oxidative Injury** section for full listing on page 40

Latanoprost EIA Kit 516811

17-phenyl-13,14-dihydro trinor $PGF_{2\alpha}$ isopropyl ester, PhXA 85
Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 58 pg/ml (free acid); 200 pg/ml (ester)
80% B/B_0: 15 pg/ml (free acid); 43 pg/ml (ester)
Summary: Latanoprost is an F-series PG analog which has been approved for use as an ocular hypotensive drug. PG esters act as prodrugs which are converted to the active free acid form by an esterase/amidase activity in ocular tissues. The Cayman Latanoprost EIA is a sensitive method for measuring the free acid form of Latanoprost. It also detects the isopropyl ester form of Latanoprost, but with less sensitivity. The assay is most appropriate for use when only one of the two forms is present.

96 wells
480 wells

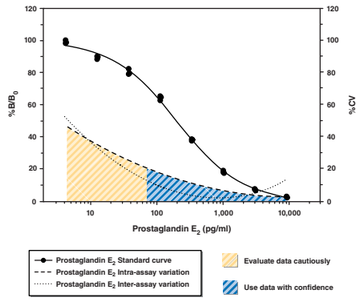


• Also Available: **Latanoprost EIA Kit (Solid Plate)** (516811.1)

Luminex® Prostaglandin E_2 Kit 10007501

Stability: ≥1 year at -20°C
Summary: Cayman's Luminex® PGE_2 is the first of its kind for the measurement of PGE_2 using Luminex® xMAP® technology. For this application microspheres have been coated with Cayman's high-affinity PGE_2 monoclonal antibody. The assay is based on the competition between PGE_2 and a PGE_2-phycoerytherin conjugate (PGE_2 tracer) for the monoclonal antibody binding sites on the microsphere beads.

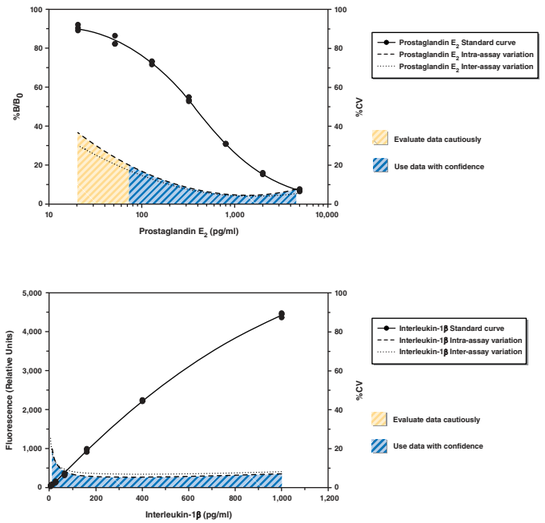
1 ea



Luminex® Prostaglandin E_2/ Interleukin-1β Duplex Kit 10009597

Stability: ≥1 year at -20°C
Summary: PGE_2 and IL-1β are inflammatory mediators that often co-exist both *in vivo* and *in vitro*. Cayman's PGE_2-IL-1β Luminex® assay allows users to measure PGE_2 and IL-1β simultaneously for the first time. The unique feature of this assay is the combination of a 'sandwich'-type assay for IL-1β and a competitive assay for PGE_2. The assay requires no wash steps and can be completed in about six hours.

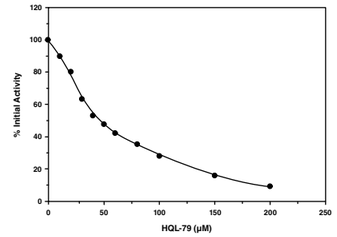
1 ea



Prostaglandin D Synthase Inhibitor Screening Assay Kit 10006595

Stability: ≥6 months at -80°C
Summary: Cayman's PGDS Inhibitor Screening Assay is a complete package for the evaluation of PGDS isozyme-specific inhibitors. Each kit includes highly-purified H-PGDS and L-PGDS, as well as PGH_2 which serves as the enzyme substrate. A complete EIA for the direct quantification of PGD_2 without prior methoximation is included in the kit.

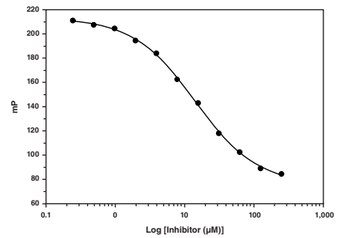
96 wells



Prostaglandin D Synthase (hematopoietic-type) FP-Based Inhibitor Screening Assay Kit - Green 600007

H-PGDS
Stability: ≥6 months at -20°C
Summary: PGD_2 is synthesized by H-PGDS in mast cells and is released in large quantities during allergic and asthmatic anaphylaxis. Cayman's H-PGDS FP-Based Inhibitor Screening Assay provides a rapid method for screening H-PGDS inhibitors. In this assay, a H-PGDS inhibitor-fluorescein conjugate serves as a specific fluorescent probe for the enzyme. Displacement of the probe by any unlabeled H-PGDS inhibitor leads to a decrease in the FP state of the probe, providing a direct signal for binding of the inhibitor to the active site of the enzyme.

384 wells
1,920 wells

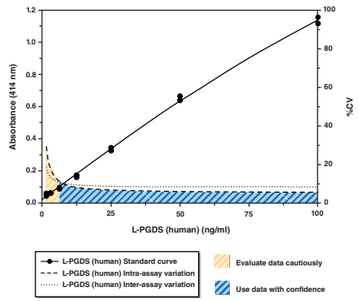


EXCLUSIVE to CAYMAN

Prostaglandin D Synthase (lipocalin-type; human) EIA Kit 10007684

L-PGDS EIA Kit, Lipocalin-PGDS ELISA Kit
Stability: ≥6 months at -20°C **Limit of Detection:** 6 ng/ml
Summary: Lipocalin-type PGDS (a.k.a., β-trace) has two functions: it catalyzes the conversion of PGH_2 to PGD_2 and acts as a carrier protein for lipid-like molecules (*i.e.*, retinoids and thyroid hormones). L-PGDS is present in a variety of body fluids including cerebrospinal fluid, seminal fluid, and plasma. This assay has been validated using CSF which contains approximately 12-30 µg/ml of L-PGDS.

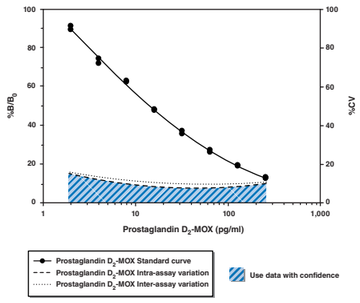
96 wells
480 wells



Prostaglandin D_2 EIA Kit 512021

Stability: ≥1 year at -80°C
Sensitivity: 50% B/B_0: 840 pg/ml • 80% B/B_0: 200 pg/ml
Summary: The direct measurement of PGD_2 in an EIA format is possible with Cayman's PGD_2 EIA Kit. The antibody utilized in this assay was generated in a unique way allowing the direct measure of PGD_2 without prior conversion to the methoximine compound, as required in our PGD_2-MOX and PGD_2-MOX Express EIA Kits (Catalog Nos. 512011 and 500151). The assay has been validated specifically for PGD_2 measurements from tissue culture supernatants or purified enzyme preparations.

96 wells
480 wells

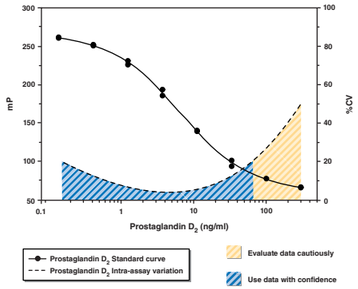


•Also Available: **Prostaglandin D_2 EIA Kit (Solid Plate)** (10006693)

Prostaglandin D_2 FPIA Kit - Red 10007835

PGD_2 Fluorescence Polarization Immunoassay - Red
Stability: ≥1 year at -80°C **Limit of Detection:** 400 pg/ml **Z' Factor:** 0.74
Summary: Cayman's PGD_2 FPIA is espically deisgned for HTS applications for direct, rapid measurement of PGD_2. The assay is particularly suited for samples from cell culture and purified enzyme preparations.

384 wells
1,920 wells

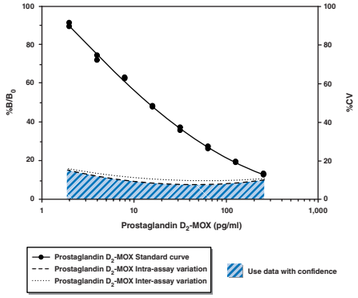


•Also Available: **Prostaglandin D_2 FPIA Kit - Green** (500581)

Prostaglandin D_2-MOX EIA Kit 512011

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 15 pg/ml • 80% B/B_0: 3.1 pg/ml
Summary: This PGD_2-MOX EIA is based on the conversion of PGD_2 to a stable MOX derivative. Treatment of the sample with MOX HCl converts PGD_2 into PGD_2-MOX, preventing its further chemical degradation.

96 wells
480 wells



•Also Available: **Prostaglandin D_2-MOX EIA Kit (Solid Plate)** (512011.1)

Olivia May, Ph.D.

Determining Cell Vitality

Measurements for cell viability are used to evaluate the death or life of cancerous cells, the rejection of implanted organs, or the effectiveness of a drug candidate. Cell viability assays are also often useful to determine optimal growth conditions of cell populations maintained in culture. Cayman offers a group of related assays that can be used to assess proliferative activity, cell viability, metabolic activity, cell cycle phase, cell toxicity, and apoptosis. Together, the information derived from these assays can indicate whether a cell population that has been exposed to an experimental stimulus is healthy or dying, actively dividing or in stasis, or has committed to an apoptotic pathway. Like a detective searching for clues, in order to gather proper evidence, it's important to ask the right questions, or in a researcher's case, select the most appropriate assay. With the variety of assay approaches available, this task can seem daunting. Luckily, features of the very cells themselves provide signs of proliferative activity and capability. Here's how to read them:

The most direct means of measuring cell proliferation, a determination of the number of actively dividing cells, is to count the number of cells present. Cell viability, defined as the number of healthy cells in a sample, determines the amount of cells (regardless of phase around the cell cycle) that are living or dead, based on a total cell sample. While a basic cell count is a direct measure of proliferation and viability, measurements of DNA content or metabolic activity are correlates that can offer more information about the physical condition and cell cycle stage.

Plasma Membrane Integrity

Assessing cell membrane integrity is one of the most common and straightforward ways to measure cell viability and assess cytotoxic consequences. Compounds that have cytotoxic effects often compromise cell membrane integrity and induce necrosis (see side bar). Dyes, such as propidium iodide, are normally excluded from the inside of healthy cells; however, if the cell membrane has been compromised, they freely cross the membrane and stain intracellular components. This method distinguishes healthy cells with uncompromised membrane integrity (unlabeled) from non-healthy ones (colored). Likewise, the labeling component of Cayman's **7-AAD Cell Viability Assay** is excluded from live cells but penetrates leaky membranes of dead or damaged cells to label the DNA. The concept of this assay relies upon the ability of 7-amino-Actinomycin D to insert itself between the tops of successive cytosine/ guanine bases of the DNA double strand, when the interior of the cell and the nuclear chromatin are accessible.

Alternatively, membrane integrity can be assessed by monitoring the passage of substances that are normally sequestered inside cells to the extracellular environment. One commonly measured molecule is lactate dehydrogenase (LDH), a soluble cytosolic enzyme that is released into the culture medium following loss of membrane integrity. Cayman's **LDH Cytotoxicity Assay** detects this cytosolic enzyme released as a result of membrane breakdown during necrosis.

Healthy, intact cells can also be fixed, detergent-permeabilized, and similarly labeled as above to study cell cycle progression. Propidium iodide, used in Cayman's **Cell Cycle Phase Determination Assay**, labels DNA in cells undergoing various phases of the cell cycle. Because the dye directly intercalates with the base pairs on a DNA strand, its fluorescent intensity is directly proportional to the DNA content of the cell. Based on chromosome distribution, it can reliably indicate G_0/G_1 *verus* S *verus* G_2/M. Further, the **CFSE Cell Division Assa**y employs a fluorescent dye that is capable of diffusing through the plasma membranes of healthy cells. As the cell divides, the fluorescence is retained in successive generations, though sequentially decreases by half with each division of c**ells. A companion kit,** 7-AAD/CFSE Cell-Mediated Cytotoxicity Assay Kit, identifies proliferating (dividing) cells, while concomitantly marking the DNA of damaged or dead cells.

Mitochondrial Activity and Apoptosis

While the most prominent role for mitochondria is the production of ATP, the major source of cellular energy, these power generators have been linked to a full gamut of cellular activities. Other important roles of mitochondria include cell signaling, cellular differentiation, cell death, as well as the control of the cell cycle and cell growth. Given these collective functions, assessing mitochondrial activity gives a fair indication of cell health. Often mitochondrial damage accompanies cytotoxic effects. Also, mitochondria can trigger apoptosis by disrupting electron transport, oxidative phosphorylation, and ATP production, by releasing proteins that activate caspase family proteases, or by altering cellular oxidation-reduction potential. JC-1 is a carbocyanine liquid crystal-forming dye used to analyze mitochondrial membrane potential. Cayman's **JC-1 Mitochondrial Membrane Potential Assay** reliably probes mitochondrial membrane potential changes occurring specifically in the early stages of apoptosis. Cayman's Caspase-3 Fluorescence Assay detects mitochondrial-triggered apoptosis by identifying activation of the specific apoptotic marker, caspase-3.

Additional assay kits available from Cayman can be used to detect morphological changes related to distinct time points during the apoptotic process. The **Annexin V FITC Assay Kit** reveals changes in plasma membrane asymmetry that signify early stage apoptosis. Phospholipids are asymmetrically distributed at the plasma membrane with phosphatidylserine (PS) predominantly observed on the inner surface facing the cytosol. In the early phases of apoptosis, though the cell membrane remains intact, this asymmetry is disrupted, thereby exposing PS to the outer layer of the membrane. Annexin V (in the presence of Ca^{2+}) preferentially binds to negatively charged phospholipids like PS. End stages of apoptosis can be assessed using Cayman's **Apoptotic Blebs Assay**, which utilizes an autoantibody variant that binds to the autoantigen in membrane blebs of apoptotic cells.

Metabolic Activity

Many researchers accept metabolic activity as an alternative for measuring proliferation. While cell number is an absolute measure of cell proliferation, metabolic activity is more a measure of cell health. Combining these assays can yield a more detailed view of cell activity. In cancer drug discovery, for example, the goal may not be to cause cell death but simply to knock down the metabolic and proliferative activity of a cell with cell stasis being the desired outcome.

A classic approach to assessing metabolic activity involves the use of tetrazolium salts that are cleaved in the mitochondria of metabolically active cells to form colored, water-insoluble (MTT) or water-soluble (XTT, WST-1, WST-8) formazan salts that can be measured by absorbance (see Figure 1).The amount of formazan dye produced is directly proportional to the number of metabolically active cells and indicates the reducing potential of the cell. The MTT (method of transcriptional and translational) assay has a long-held reputation as the conventional cell proliferation assay and carries with it a strong body of literature support. However, the MTT assay is not as sensitive as newer tetrazolium salt assays. Another drawback is the fact it is cleaved to a water-insoluble formazan crystal that must be solubilized before reading on the spectrophotometer, which adds an additional step to the assay protocol.

XTT, WST-1, and WST-8 produce aqueous-soluble formazan products, eliminating the need for a solubilization step, which facilitates assay optimization. In particular, WST-8 is more stable and less cytotoxic compared to other tetrazolium salts, making it especially useful for longer incubation periods. Furthermore, the detection sensitivity of WST-8 is higher than that for other tetrazolium salts.

In comparison to traditional, radioactive [^{3}H]-thymidine or nonradioactive 5'-bromo-2'deoxy-uridine (BrdU) assays, MTT, XTT, WST-1, and WST-8 have the advantage of ease of use. Precautions, though, must be considered with data interpretation when using tetrazolium salts for cell quantitation as important data may invariably be excluded. The very nature of the tetrazolium dye interferes with cell metabolism, and the changes that result may or may not indicate whether the cell is viable and proliferating. Since the basis of the assay is the inherent dehydrogenase activity of viable cells, treatments that affect dehydrogenase activity may result in a discrepancy between the actual viable cell number and that determined using the formazan dye.

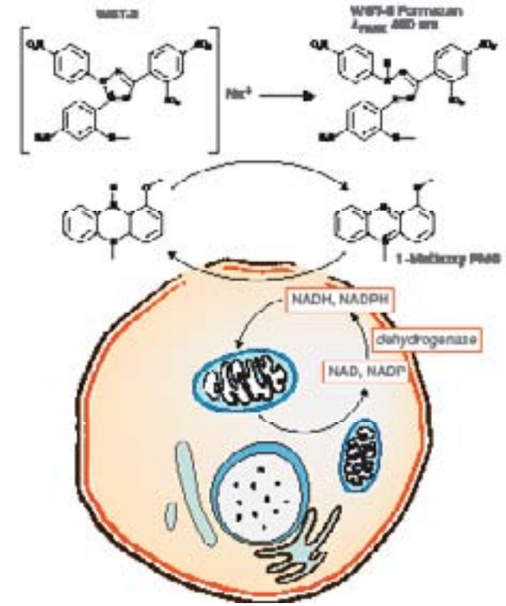
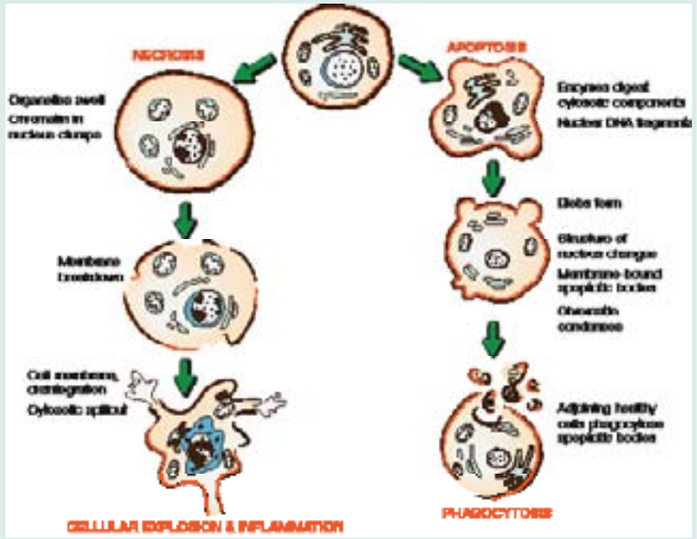


Figure 1. Reactive oxygen species generated by NADPH oxidase

CELL DEATH

Compromised cells may undergo necrosis, in which they lose membrane integrity and die rapidly as a result of cell lysis, they can stop actively growing and dividing (a decrease in cell viability), or the cells can activate a genetic program of controlled cell death (apoptosis). Cells undergoing necrosis typically exhibit rapid swelling, lose membrane integrity, shut down metabolism, and release their contents into the environment, which triggers an inflammatory response. Cells that undergo rapid necrosis in vitro do not have adequate time or energy to activate apoptotic machinery and will not express apoptotic markers such as caspase.

On the other hand, apoptosis is characterized by controlled cytological and molecular events including a change in the refractive index of the cell, cytoplasmic shrinkage, and overall decreases in cell volume, nuclear condensation, and cleavage of DNA into regularly sized fragments. The apoptotic cell divides into many parts by "blebbing" and "budding", to form what are known as 'apoptotic bodies', containing cell organelles and nuclear materials surrounded by an intact plasma membrane. Eventually, these bodies are phagocytosed by neighboring healthy cells and recycled.

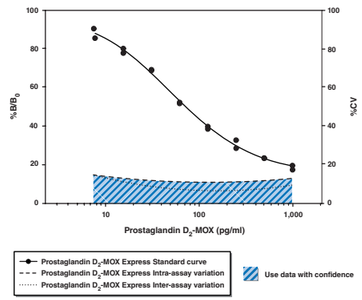


Which Kit is right for YOU?	Proliferation	Viability	Cytotoxicity	Apoptosis
Cell Cycle Phase Determination Kit	✓	✓		
7-AAD Cell Viability Assay Kit		✓	✓	
CFSE Cell Division Assay Kit	✓			
7-AAD/CFSE Cell-Mediated Cytotoxicity Assay Kit	✓	✓	✓	
LDH Cytotoxicity Assay Kit		✓	✓	
JC-1 Mitochondrial Membrane Potential Assay Kit				✓
Caspase-3 Fluorescence Assay Kit				✓
Apoptotic Blebs Assay				✓
MTT Cell Proliferation Assay Kit	✓	✓	✓	
XTT Cell Proliferation Assay Kit	✓	✓	✓	
WST-1 Cell Proliferation Assay Kit	✓	✓	✓	
WST-8 Cell Proliferation Assay Kit	✓	✓	✓	

Prostaglandin D_2-MOX Express EIA Kit 500151

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 75 pg/ml • 80% B/B_0: 16 pg/ml
Summary: Cayman's PGD_2-MOX Express EIA is a competitive assay that permits the rapid measurement of PGD_2 from biological samples, requiring only one hour incubation and development times for each step.

96 wells
480 wells

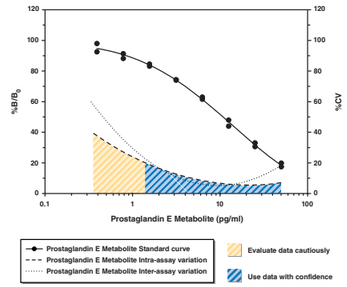


•Also Available: **Prostaglandin D_2-MOX Express EIA Kit (Solid Plate)** (500151.1)

Prostaglandin E Metabolite EIA Kit 514531

PGEM
Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 11 pg/ml • 80% B/B_0: 2 pg/ml
Summary: Because of the rapid metabolism of PGE_2, the determination of *in vivo* PGE_2 biosynthesis is often best accomplished by the measurement of PGE_2 metabolites. Cayman's PGEM Assay converts all 13,14-dihydro-15-keto PGE_2 and 13,14-dihydro-15-keto PGA_2 into a single stable derivative, which is easily measurable by EIA. This assay is therefore the method of choice if the samples in question have undergone extensive metabolism prior to collection.

96 wells
480 wells



•Also Available: **Prostaglandin E Metabolite EIA Kit (Solid Plate)** (514531.1)

Cyclooxygenase (COX) Activity and Inhibitor Screening Assays

Kit	Detection Method	Activity Measured	Reagents/Equipment	Interferences	Samples Assayed	Format/ Time	Stability
COX Activity Assay Kit cat. no. 760151	Peroxidase activity is assayed colorimetrically by monitoring the appearance of oxidized TMPD. The assay includes COX-1 and COX-2 specifc inhibitors in order to distinguish between the two enzymes.	Peroxidase activity	Everything is included. **Plate reader with 590-611 nm filter is required.**	Antioxidants	13 samples in duplicate including background value, sample value, and one inhibitor for identifying COX isozyme present in the sample.	Reactions performed in 96-well plate (answer in 30 minutes)	1 year at -80°C
COX Fluorescent Activity Assay Kit cat. no. 700200	Fluorometric--monitors the conversion of ADHP to resorufin	Peroxidase activity	Fluorometer with capacity to measure fluorescence using an excitation wavelength of 530-540 nm and emission wavelength of 585-595 nm is required.	Triton-X-100 (1%) and Glutathione (1 mM)	30 samples in triplicate or 46 in duplicate	Reactions performed in 96-well plate (answer in 30 minutes)	1 year at -80°C
COX Fluorescent Inhibitor Screening Assay Kit cat. no. 700100	Fluorescence-based method for screening COX-1 and COX-2 isozyme-specific inhibitors	Peroxidase activity of ovine COX-1 and human recombinant COX-2	Fluorometer with capacity to measure fluorescence using an excitation wavelength of 530-540 nm and emission wavelength of 585-595 nm is required.	Antioxidants	30 samples in triplicate or 46 in duplicate	Reactions performed in 96-well plate (answer in 30 minutes)	1 year at -80°C
COX (ovine) Inhibitor Screening Assay Kit cat. no. 560101	PGs generated in COX reaction are quantified by EIA.	COX and peroxidase activities of ovine COX-1 and ovine COX-2	Everything is included except inhibitors of interest. **Water bath and plate reader with 405-414 nm filter is required.**	Inhibitors may interfere with EIA. Samples may have to be purified before performing EIA.	36 samples in duplicate	Generate PGs in COX rxn, then perform EIA (answer in 2 days)	1 year at -80°C
COX Inhibitor Screening Assay Kit cat. no. 560131	PGs generated in COX reaction are quantified by EIA.	COX and peroxidase activities of ovine COX-1 and human recombinant COX-2	Everything is included except inhibitors of interest. **Water bath and plate reader with 405-414 nm filter is required.**	Inhibitors may interfere with EIA. Samples may have to be purified before performing EIA.	36 samples in duplicate	Generate PGs in COX rxn, then perform EIA (answer in 2 days)	1 year at -80°C
Colorimetric COX (ovine) Inhibitor Screening Assay Kit cat. no. 760111	Peroxidase activity is assayed colorimetrically by monitoring the appearance of oxidized TMPD.	Peroxidase activity	Everything is included except inhibitors of interest. **Plate reader with 590-611 nm filter is required.**	Antioxidants	48 samples in duplicate	Reactions performed in 96-well plate (answer in 30 minutes)	1 year at -80°C

Cayman Chemical's Inhibitor Screening Assays are used to screen for isozyme-specific inhibitors. All kits contain both COX-1 and COX-2. The COX Activity Assays can be used to detect COX-activity in cell lysates, tissue homogenates, and purified enzyme preparations.

Prostaglandin E_2 Affinity Purification Kit 514018

Stability: ≥2 years at 4°C
Summary: This kit contains all reagents necessary for a simple one-step purification of most biological samples.

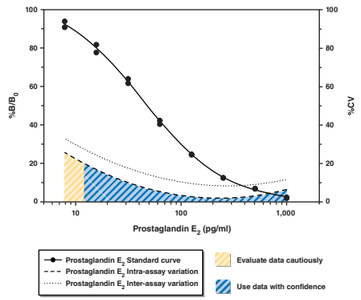
1 ea
5 ea

•Also Available: **Prostaglandin E_2 Affinity Column** (414018)
Prostaglandin E_2 Affinity Sorbent (414020)

Prostaglandin E_2 EIA Kit - Monoclonal 514010

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 50 pg/ml • 80% B/B_0: 15 pg/ml
Summary: Cayman's PGE_2 EIA is a sensitive competitive assay that uses a high-affinity PGE_2 monoclonal antibody for quantification of PGE_2 in plasma, urine, and culture media samples.

96 wells
480 wells

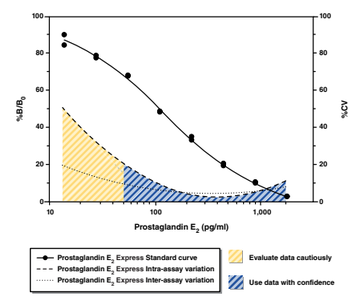


•Also Available: **Prostaglandin E_2 EIA Kit - Monoclonal (Solid Plate)** (514010.1)

Prostaglandin E_2 Express EIA Kit 500141

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 125 pg/ml • 80% B/B_0: 36 pg/ml
Summary: Cayman's PGE_2 Express EIA is a competitive assay that permits the rapid measurement of PGE_2 from biological samples, requiring only one hour incubation and development times for each step.

96 wells
480 wells

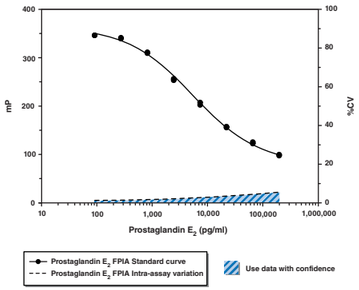


•Also Available: **Prostaglandin E_2 Express EIA Kit (Solid Plate)** (500141.1)

Prostaglandin E_2 FPIA Kit - Red 10004517

PGE_2 Fluorescence Polarization Immunoassay - Red
Stability: ≥6 months at -20°C **Limit of Detection:** 100 pg/ml **Z' Factor:** 0.69
Summary: Cayman's PGE_2 FPIA is an excellent method for rapid, high-throughput screening of PGE_2 samples. This assay utilizes the red-shifted dye rhodamine as the label.

384 wells
1,920 wells

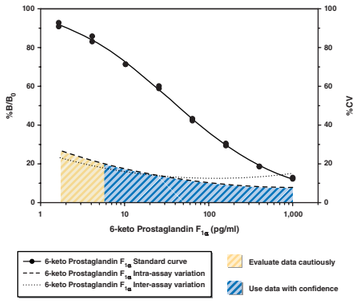


•Also Available: **Prostaglandin E_2 FPIA Kit - Green** (500501)

6-keto Prostaglandin $F_{1\alpha}$ EIA Kit 515211

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 40 pg/ml • 80% B/B_0: 6 pg/ml
Summary: Prostacyclin is formed from arachidonic acid primarily by the vascular endothelium and renal cortex. It is a potent vasodilator and inhibitor of platelet aggregation. PGI_2 is non-enzymatically hydrated to 6-keto $PGF_{1\alpha}$ ($t_{1/2}$ = 2-3 minutes), and then quickly converted to the major metabolite, 2,3-dinor-6-keto $PGF_{1\alpha}$ ($t_{1/2}$ = 30 minutes).

96 wells
480 wells

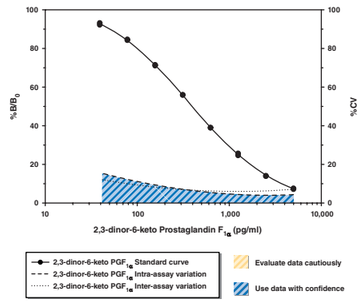


•Also Available: **6-keto Prostaglandin $F_{1\alpha}$ EIA Kit (Solid Plate)** (515211.1)

2,3-dinor-6-keto Prostaglandin $F_{1\alpha}$ EIA Kit 515121

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 400 pg/ml • 80% B/B_0: 100 pg/ml
Summary: PGI_2 is non-enzymatically hydrated to 6-keto $PGF_{1\alpha}$, and then quickly converted to the major urinary metabolite, 2,3-dinor-6-keto $PGF_{1\alpha}$. The majority of 6-keto $PGF_{1\alpha}$ in urine is of renal origin with only 14% originating from plasma. Cayman's 2,3-dinor-6-keto $PGF_{1\alpha}$ EIA utilizes a highly selective monoclonal antibody that exhibits no cross reactivity with 6-keto $PGF_{1\alpha}$, thus providing a method for accurate measurement of systemic PGI_2 production.

96 wells
480 wells

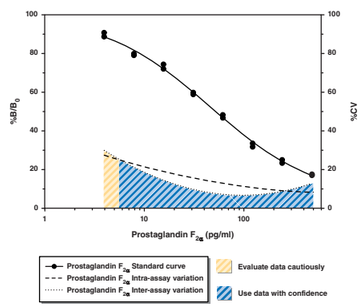


•Also Available: **2,3-dinor-6-keto Prostaglandin $F_{1\alpha}$ EIA Kit (Solid Plate)** (10008826)

Prostaglandin $F_{2\alpha}$ EIA Kit 516011

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 52 pg/ml • 80% B/B_0: 9 pg/ml
Summary: $PGF_{2\alpha}$ is one of the five primary PGs derived enzymatically directly from the endoperoxide PGH_2. The majority of the functional roles ascribed to it relate to fertility, pregnancy, and parturition. Like all of the primary PGs, $PGF_{2\alpha}$ has a very short half-life in the general circulation. The plasma concentration of $PGF_{2\alpha}$ in humans is <10 pg/ml, and probably no more than 1-2 pg/ml.

96 wells
480 wells

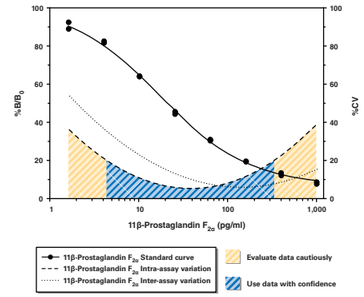


•Also Available: **Prostaglandin $F_{2\alpha}$ EIA Kit (Solid Plate)** (516011.1)

11β-Prostaglandin $F_{2\alpha}$ EIA Kit 516521

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 32 pg/ml • 80% B/B_0: 5.5 pg/ml
Summary: 11β-$PGF_{2\alpha}$ is the primary plasma metabolite of PGD_2 *in vivo*, the levels of which can increase from 6 pg/ml in a normal healthy volunteer to 490 ng/ml in patients with systemic mastocytosis.

96 wells
480 wells

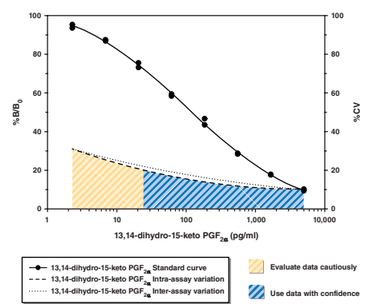


•Also Available: **11β-Prostaglandin $F_{2\alpha}$ EIA Kit (Solid Plate)** (516521.1)

13,14-dihydro-15-keto Prostaglandin $F_{2\alpha}$ EIA Kit 516671

PGFM
Stability: ≥6 months at -20°C
Sensitivity: 50% B/B_0: 120 pg/ml • 80% B/B_0: 13 pg/ml
Summary: $PGF_{2\alpha}$ is rapidly metabolized to 13,14-dihydro-15-keto $PGF_{2\alpha}$ *in vivo*, by the enzymes 15-hydroxy PG dehydrogenase and Δ^{13}-reductase. Measurement of 13,14-dihydro-15-keto $PGF_{2\alpha}$ in plasma can be used as a marker of the *in vivo* production of $PGF_{2\alpha}$. The assay has been validated for the measurement of 13,14-dihydro-15-keto $PGF_{2\alpha}$ in plasma, a common matrix for measurement of this $PGF_{2\alpha}$ metabolite.

96 wells
480 wells

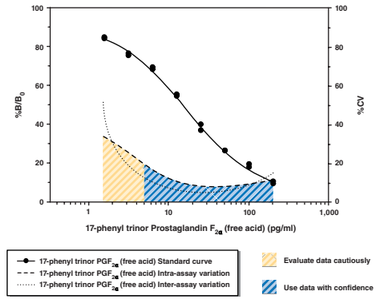


•Also Available: **13,14-dihydro-15-keto Prostaglandin $F_{2\alpha}$ EIA Kit (Solid Plate)** (516671.1)

17-phenyl trinor Prostaglandin $F_{2\alpha}$ EIA Kit 516821

Bimatoprost
Stability: ≥6 months at -20°C
Sensitivity: 50% B/B_0: 15.5 pg/ml • 80% B/B_0: 2.6 pg/ml
Summary: 17-phenyl trinor $PGF_{2\alpha}$ is a metabolically stable analog of $PGF_{2\alpha}$ and is a potent agonist for the FP receptor. The ethyl amide analog has been approved for use as an ocular hypotensive drug, sold under the Allergan trade name Bimatoprost. Cayman's 17-phenyl trinor $PGF_{2\alpha}$ EIA is a sensitive detection method for measuring both the free acid and ethyl amide forms of 17-phenyl trinor $PGF_{2\alpha}$. The assay is most appropriate for use when only one of the two forms is present.

96 wells
480 wells



•Also Available: **17-phenyl trinor Prostaglandin $F_{2\alpha}$ (Solid Plate)** (516821.1)

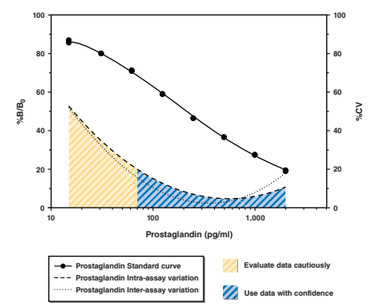
ent-Prostaglandin $F_{2\alpha}$ EIA Kit 10010382

Please see the **Oxidative Injury** section for full listing on page 42

Prostaglandin Screening EIA Kit 514012

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 220 pg/ml • 80% B/B_0: 29 pg/ml
Summary: This assay was developed for screening applications in which the relative amount of PG production for a large number of cell culture samples must be determined. The antiserum used in this assay exhibits high cross reactivity for most PGs which will allow quantification of all the PGs in a given sample with a single assay.

96 wells
480 wells

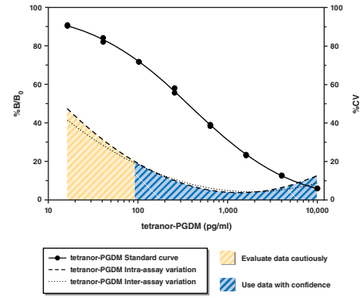


•Also Available: **Prostaglandin Screening EIA Kit (Solid Plate)** (514012.1)

EXCLUSIVE to CAYMAN
tetranor-PGDM EIA Kit 501001

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 300 pg/ml • 80% B/B_0: 55 pg/ml
Summary: tetranor-PGDM is a major metabolite of PGD_2 found in human and murine urine with normal levels of 1.5 ng/mg creatinine and 8.1 ng/mg creatinine respectively. Cayman's tetranor-PGDM EIA is a competitive assay that can be used for quantification of tetranor-PGDM in urine.

96 wells
480 wells



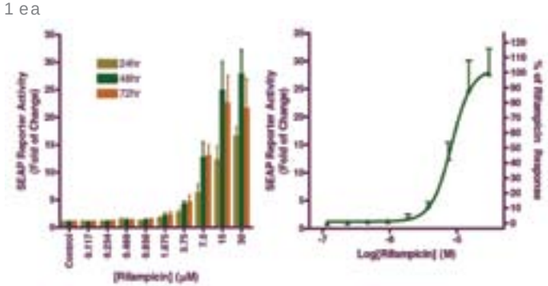
•Also Available: **tetranor-PGDM EIA Kit (Solid Plate)** (501002)

STEP Reporter Assays

CYP3A4 Induction STEP Reporter Assay Kit (Luminescence) 600270

Stability: ≥1 year at -20°C
Summary: Cayman's CYP3A4 Induction STEP Reporter Assay (Luminescence) is a novel method to assess CYP3A4 induction. STEP (Surface Transfection and Expression Protocol) is a patented, solid phase transient transfection technology that overcomes many of the disadvantages of other transfection approaches. Cayman's assay consists of a 96-well plate coated with a STEP complex containing a Secreted Alkaline Phosphatase (SEAP) reporter regulated by the human CYP3A4 gene promoter. The STEP complex also contains two nuclear expression constructs, Pregnane X Receptor (PXR) and Hepatocyte Nuclear Factor-4α (HNF-4α). Cells grown on the CYP3A4 STEP plate will introduce the reporter gene and express PXR and HNF-4α. Binding of inducer-activated transcription factors to the CYP3A4 promoter initiates expression of SEAP, which is secreted into the cell culture medium. Aliquots of medium are removed at fixed time intervals and SEAP activity is measured by adding a luminescence-based alkaline phosphatase substrate provided in the kit. The kit is simple to use and can be easily adapted to HTS for potential CYP3A4 inducers.

1 ea



Melanocortin 3 Receptor STEP Reporter Assay Kit (Luminescence) 600180

MC3R
Stability: ≥1 year at -20°C
Summary: The melanocortin-3 receptor (MC3R) helps regulate energy homeostasis and mice lacking MC3R have increased fat mass and reduced lean mass. Therefore, agonists that selectively activate MC3R might have beneficial effects related to weight gain and glucose metabolism. This assay consists of a 96-well plate coated with both MC3R and Secretory Alkaline Phosphatase (SEAP) reporter constructs. Cells grown on the STEP complex will express MC3R at the cell surface. Binding of agonists to MC3R initiates a signaling cascade resulting in expression of SEAP. SEAP activity is measured following addition of a luminescence-based alkaline phosphatase substrate provided in the kit.

1 ea

Melanocortin 4 Receptor STEP Reporter Assay Kit (Luminescence) 600190

MC4R
Stability: ≥1 year at -20°C **Summary:** The melanocortin-4 receptor (MC4R) has important roles in weight regulation, sexual function, and inflammation. Mice deficient in MC4R have increased lipid deposition associated with elevated adiposity, while mutations in MC4R in humans are associated with early onset or severe obesity. This assay consists of a 96-well plate coated with both MC4R and Secretory Alkaline Phosphatase (SEAP) reporter constructs. Cells grown on the STEP complex will express MC4R at the cell surface. Binding of agonists to MC4R initiates a signaling cascade resulting in expression of SEAP. SEAP activity is measured following addition of a luminescence-based alkaline phosphatase substrate provided in the kit.

1 ea

Orexin 1 Receptor STEP Reporter Assay Kit (Luminescence) 600240

OX1R
Stability: ≥1 year at -80°C
Summary: The Orexin 1 Receptor (OX1R) may be an important therapeutic target for treatment of sleep disorders, obesity, emotional stress, and addiction. This assay consists of a 96-well plate coated with both OX1R and Secretory Alkaline Phosphatase (SEAP) reporter constructs. Cells grown on the STEP complex will express OX1R at the cell surface. Binding of agonists to OX1R initiates a signaling cascade resulting in expression of SEAP. SEAP activity is measured following addition of a luminescence-based alkaline phosphatase substrate provided in the kit.

1 ea

Orexin 2 Receptor STEP Reporter Assay Kit (Luminescence) 600250

OX2R
Stability: ≥1 year at -80°C
Summary: The Orexin 2 Receptor (OX2R) may be an important therapeutic target for treatment of sleep disorders, obesity, emotional stress, and addiction. This assay consists of a 96-well plate coated with both OX2R and Secretory Alkaline Phosphatase (SEAP) reporter constructs. Cells grown on the STEP complex will express OX2R at the cell surface. Binding of agonists to OX2R initiates a signaling cascade resulting in expression of SEAP. SEAP activity is measured following addition of a luminescence-based alkaline phosphatase substrate provided in the kit.

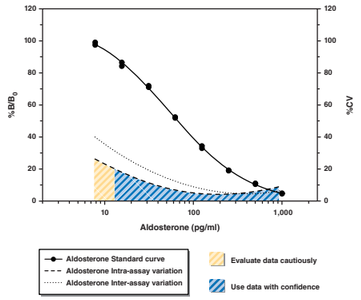
1 ea

Steroids

Aldosterone EIA Kit - Monoclonal 10004377

Stability: ≥6 months at -20°C
Sensitivity: 50% B/B_0: 65 pg/ml • 80% B/B_0: 21 pg/ml
Summary: Aldosterone, a steroid hormone secreted by the adrenal cortex, is the principle mineralocorticoid controlling sodium and potassium balance. The primary role of aldosterone is to promote unidirectional salt reabsorption across a variety of epithelial tissues, the salivary gland, intestine, sweat glands, and the kidney.

96 wells
480 wells

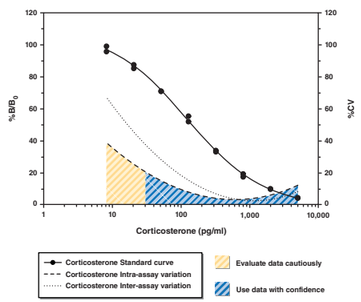


•Also Available: **Aldosterone EIA Kit - Monoclonal (Solid Plate)** (10004553)

Corticosterone EIA Kit 500655

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 150 pg/ml • 80% B/B_0: 30 pg/ml
Summary: Corticosterone is a glucocorticoid produced by the adrenal cortex in response to adrenocorticotropic hormone, and is the precursor to aldosterone. The production of glucorcorticoids is increased by stress; therefore, corticosterone can be used as a biomarker of stress. Cayman's Corticosterone EIA is a competitive assay that has been validated for the measurement of corticosterone from plasma and fecal samples.

96 wells
480 wells

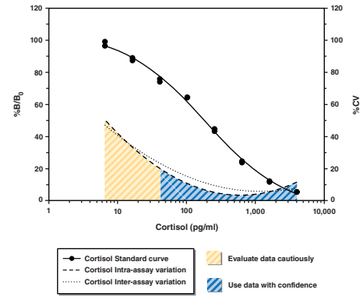


•Also Available: **Corticosterone EIA Kit (Solid Plate)** (500656)

Cortisol EIA Kit 500360

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 180 pg/ml • 80% B/B_0: 35 pg/ml
Summary: Cortisol is a glucocorticoid produced by the adrenal cortex in response to adrenocorticotropic hormone. It is secreted with a circadian periodicity, and peaks just prior to waking in the morning. Cortisol is often elevated in major depressive disorder, certain forms of hypertension, stress, AIDS, and in the visceral fat of obese individuals. Cortisol can be measured in many matrices including blood, urine, and saliva.

96 wells
480 wells



•Also Available: **Cortisol EIA Kit (Solid Plate)** (500361)

Cortisol Express EIA Kit 10006791

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 80 pg/ml • 80% B/B_0: 17 pg/ml
Summary: Cayman's Cortisol Express EIA is a competitive assay that permits the rapid measurement of cortisol from biological samples, requiring only a two hour incubation and one hour development times.

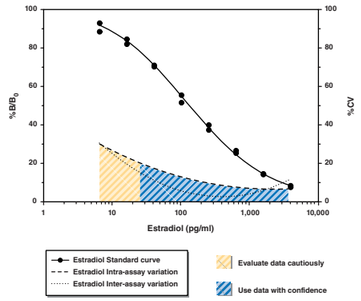
96 wells
480 wells

•Also Available: **Cortisol Express EIA Kit (Solid Plate)** (10007337)

Estradiol EIA Kit 582251

β-Estradiol, 17β-Estradiol, β-Oestradiol
Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: ~130 pg/ml • 80% B/B_0: ~19 pg/ml
Summary: Estradiol is the major estrogen secreted by the premenopausal ovary. Estrogens direct the development of the female genotype in embryogenesis and at puberty. In addition, estradiol is an important luteolytic agent in humans. Plasma estradiol levels increase gradually between days 1-7 of the menstrual cycle followed by a sharp increase to a peak value of about 300 pg/ml on day 12 just prior to ovulation.

96 wells
480 wells

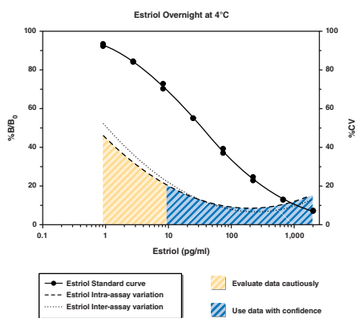


•Also Available: **Estradiol EIA Kit (Solid Plate)** (582251.1)

Estriol EIA Kit 582281

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 33 pg/ml (4°C - 18 hours incubation)
95 pg/ml (room temperature - 2 hour incubation)
80% B/B_0: 4 pg/ml (4°C - 18 hours incubation)
14 pg/ml (room temperature - 2 hour incubation)
Summary: Estriol is a metabolite of estradiol and a major estrogen produced during pregnancy. In the final weeks before parturition, estriol levels increase significantly (<10 ng/ml at 8-10 weeks gestation to approximately 150 ng/ml at week 38). The majority of the estriol synthesized in the later stages of pregnancy originates from fetal dehydroepiandrosterone sulfate (DHEAS) and serves as a direct marker of fetal adrenal gland activity.

96 wells
480 wells

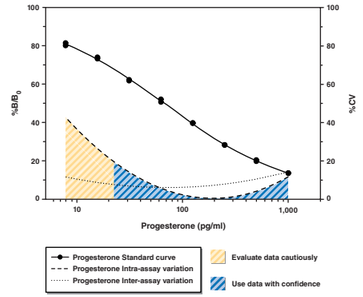


•Also Available: **Estriol EIA Kit (Solid Plate)** (582281.1)

Progesterone EIA Kit 582601

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 70 pg/ml • 80% B/B_0: 10 pg/ml
Summary: Progesterone, along with pregnenolone, is the biosynthetic precursor of all other steroid hormones. The main function of progesterone is to prepare the uterine lining for implantation of a fertilized ovum and to maintain pregnancy. Measurement of serum or plasma progesterone levels are used as an index to monitor ovulation and investigate luteal function.

96 wells
480 wells

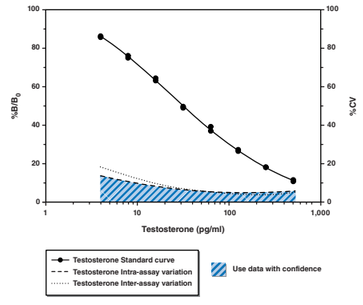


•Also Available: **Progesterone EIA Kit (Solid Plate)** (582601.1)

Testosterone EIA Kit 582701

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 32 pg/ml • 80% B/B_0: 6 pg/ml
Summary: Testosterone is the prototypic and predominant circulating androgenic steroid. It plays a major role in the growth and function of many reproductive and non-reproductive tissues and organs including muscle, liver, and brain, directing the development of the male phenotype during embryogenesis and at puberty.

96 wells
480 wells

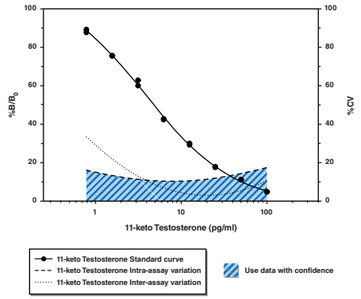


•Also Available: **Testosterone EIA Kit (Solid Plate)** (582701.1)

11-keto Testosterone EIA Kit 582751

11-KT
Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 5 pg/ml • 80% B/B_0: 1.3 pg/ml
Summary: 11-KT is a second key androgenic steroid found in fish. It occurs in males together with testosterone in amounts which vary from less than 1 ng/ml to as much as 50-100 ng/ml, depending on the species and the stage of the reproductive cycle.

96 wells
480 wells



•Also Available: **11-keto Testosterone EIA Kit (Solid Plate)** (582751.1)

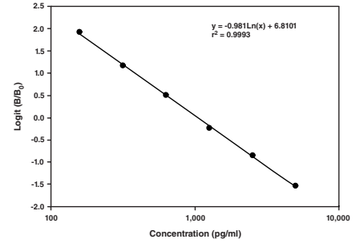
Thromboxanes

TXA_2 is produced from arachidonic acid by many cells and causes irreversible platelet aggregation and vascular and bronchial smooth muscle contraction. TXA_2 is rapidly hydrolyzed non-enzymatically to form TXB_2, which is then quickly metabolized ($t_{1/2}$ = 5-7 minutes) to urinary metabolites for clearance by the kidneys. Because of the transient nature of this compound it is difficult to accurately measure circulating levels in whole-animal experimental models. In fact, it has been shown that plasma and urine levels of TXB_2 are primarily due to *ex vivo* platelet activation and intra-renal production, respectively. Therefore, measurement of TXB_2 metabolites such as 11-dehydro TXB_2 (Catalog No. 519501) and 2,3-dinor TXB_2 (Catalog No. 519051) in urine and plasma may give better estimates of *in vivo* TXA_2 production. TXB_2 measurement is better suited towards samples that are not expected to undergo extensive metabolism such as perfusates, lavage samples, and tissue/cell culture medium or lysates.

Aspirin™ Effect-Detection Kit 10010153

Stability: ≥1 year at 4°C
Summary: The Cayman Aspirin™ Effect-Detection Kit is a 510K, clinically-approved diagnostic assay for the measurement of 11-dehydro TXB_2. It is intended to help physicians assess the effectiveness of their patients' Aspirin™ regime, and to help identify the high-risk group who are inadequately controlled on an 80 mg aspirin dose. This assay can be completed in 3 hours and utilizes urine as the sample matrix. The assay exhibits intra-assay %CV values of <11% with sensitivity sufficient to detect the lower levels of 11-dehydro TXB_2 in patients that respond well to Aspirin™.

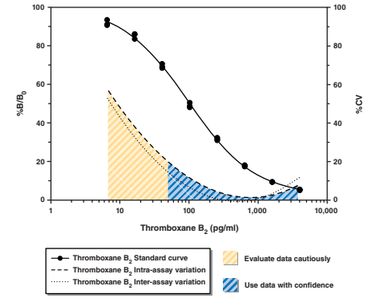
1 ea



Luminex® Thromboxane B_2 Kit 10007502

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 100 pg/ml • 80% B/B_0: 24 pg/ml

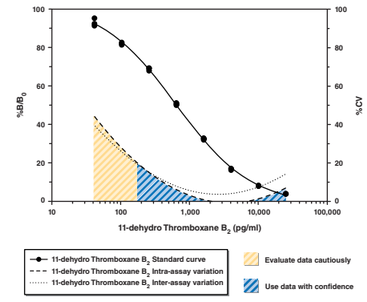
1 ea



Luminex® 11-dehydro Thromboxane B_2 Kit 10010971

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 650 pg/ml • 80% B/B_0: 130 pg/ml

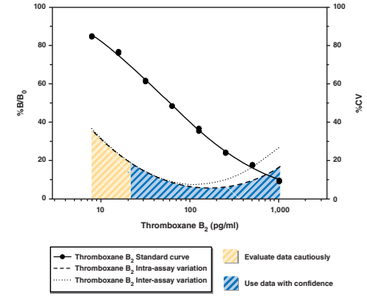
1 ea



Thromboxane B_2 EIA Kit 519031

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 57 pg/ml • 80% B/B_0: 11 pg/ml

96 wells
480 wells

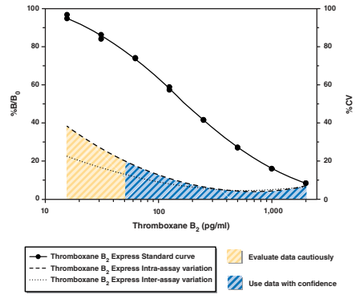


•Also Available: **Thromboxane B_2 EIA Kit (Solid Plate)** (519031.1)

Thromboxane B_2 Express EIA Kit - Monoclonal 10004023

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 176 pg/ml • 80% B/B_0: 45 pg/ml

96 wells
480 wells

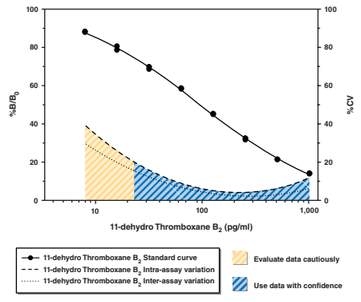


•Also Available: **Thromboxane B_2 Express EIA Kit - Monoclonal (Solid Plate)** (10005386)

11-dehydro Thromboxane B_2 EIA Kit 519501

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 93 pg/ml • 80% B/B_0: 16 pg/ml

96 wells
480 wells

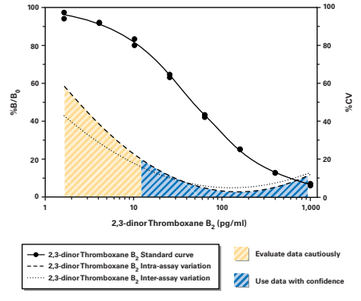


•Also Available: **11-dehydro Thromboxane B_2 EIA Kit (Solid Plate)** (519501.1)

2,3-dinor Thromboxane B_2 EIA Kit 519051

Stability: ≥1 year at -20°C
Sensitivity: 50% B/B_0: 35 pg/ml • 80% B/B_0: 7 pg/ml

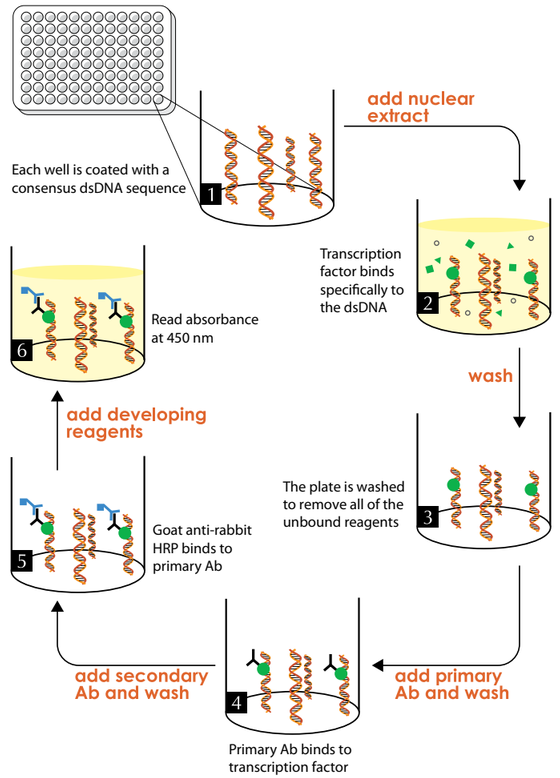
96 wells
480 wells



•Also Available: **2,3-dinor Thromboxane B_2 EIA Kit (Solid Plate)** (519051.1)

Transcription Factor Assays

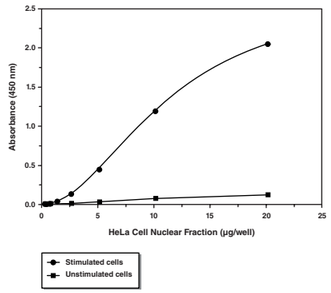
The Cayman Chemical Transcription Factor Assays are a non-radioactive, sensitive method for detecting specific transcription factor DNA binding activity in nuclear extracts. A 96-well enzyme-linked immunosorbent assay (ELISA) replaces the cumbersome radioactive electrophoretic mobility shift assay (EMSA). A specific double-stranded DNA (dsDNA) sequence containing the target-specific response element is immobilized on a 96-well plate. Transcription factors contained in a nuclear extract bind specifically to the DNA and are detected by a specific primary antibody. A secondary antibody conjugated to HRP is added to provide a sensitive colorometric readout at 450 nm.



ATF2 (Phospho-Thr[69,71]) Transcription Factor Assay Kit 600130

Activating Transcription Factor 2
Stability: ≥6 months at -80°C
Summary: ATF2 is a sequence-specific DNA-binding protein belonging to the bZIP family of transcription factors that bind with high affinity to the octameric CRE. ATF2 mediates both transcription and DNA damage control through its phosphorylation/activation in response to inflammatory cytokines, UV irradiation, alkylating compounds, and other cellular stressors.

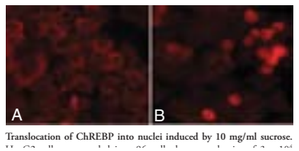
96 wells



ChREBP Cell-Based Translocation Assay Kit 10010060

Stability: ≥1 year at -20°C
Summary: The identification of ChREBP activators is of great interest for drug discovery. The distinct translocation of the protein from the cytoplasm to the nucleus during activation makes it possible to study modulators of ChREBP through sub-cellular localization of the protein using conventional immunocytochemical staining with a specific antibody. Cayman's ChREBP Cell-Based Translocation Assay provides a highly specific ChREBP primary antibody together with a Dylight™ (product of Thermo Scientific Inc.) conjugated secondary antibody in a ready to use format.

96 wells



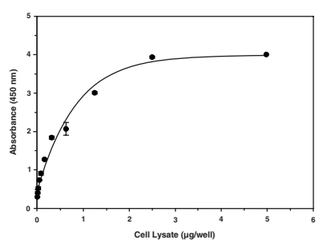
Translocation of ChREBP into nuclei induced by 10 mg/ml sucrose.

Translocation of ChREBP into nuclei induced by sucrose. *Panel A:* HepG2 cells treated with PBS (vehicle) demonstrate cytoplasmic localization of ChREBP. *Panel B:* Sucrose treatment for 24 hours induces ChREBP translocation into the nuclei.

ChREBP Transcription Factor Assay Kit 10006909

Stability: ≥6 months at -20°C
Summary: ChREBP is a transcription factor playing a critical role in the nutrient and hormonal regulation of genes encoding enzymes of glucose metabolism and lipogenesis pathways.

96 wells



CREB (Phospho-Ser[133]) Transcription Factor Assay Kit 10009846

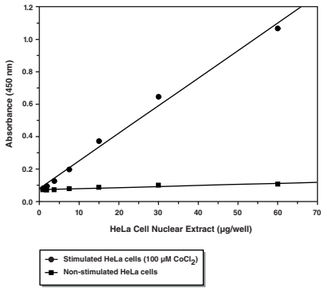
Stability: ≥6 months at -20°C
Summary: CREB (cAMP-response-element-binding protein) is a transcription factor that binds to cAMP-responsive element (CRE) promoter sites to regulate the transcription of numerous genes involved in metabolic regulation, depression, long term memory, and other physiological processes. Phosphorylation on serine 133 (Ser[133]) activates CREB to induce transcription of target genes. Diverse stimuli such as growth factors, neurotransmitters, hypoxia, growth factors, UV light, survival signals, and stress signals are some of the known activators of CREB.

96 wells

HIF-1α Transcription Factor Assay Kit 10006910

Hypoxia-inducible Factor-1α
Stability: ≥6 months at -80°C
Summary: The HIF-1α (hypoxia-inducible factor) transcription factor is a member of the basic-helix-loop-helix (bHLH) family of transcription factors and plays an important role in maintaining cellular oxygen homeostasis. HIF-1α has emerged as an important drug target in breast and prostate cancer, cardiovascular disease, and ischemia.

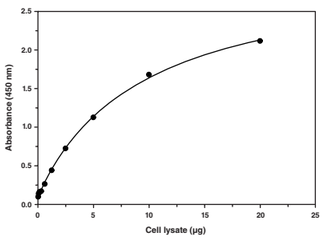
96 wells



Liver X Receptor β Transcription Factor Assay Kit 10011119

Stability: ≥1 year at -80°C
Summary: LXRs are ligand-activated transcription factors that are primarily activated by oxysterols and cholesterol metabolites. As such, LXRs play an important role in the regulation of cholesterol, lipid, and carbohydrate metabolism. There are two known isoforms of LXR: LXRα and LXRβ. LXRβ is ubiquitously expressed in all tissues while LXRα is primarily expressed in the liver, adipose tissue, small intestine, and macrophages. LXRs are currently being examined as potential therapeutic targets in the treatment of diabetes, cardiovascular disease, Alzheimers disease, obesity, and atherosclerosis.

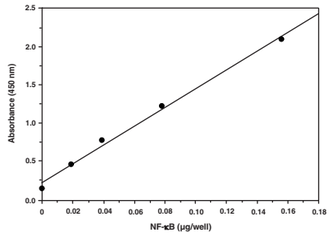
96 wells



NF-κB (human p50) Transcription Factor Assay Kit 10006912

Stability: ≥6 months at -20°C
Summary: Cayman's NF-κB (human p50) Transcription Factor Assay is a non-radioactive, sensitive mehod for detecting specific transcription factor DNA binding activity in nuclear extracts and whole cell lysates in a 96-well ELISA format. Cayman's NF-κB (human p50) Transcription Factor Assay detects NF-κB (p50). It will not cross-react with NF-κB (p65).

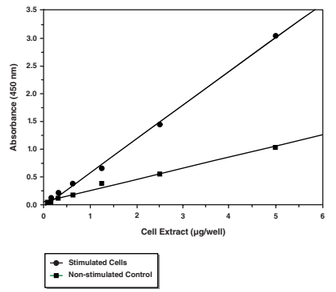
96 wells



NF-κB (p65) Transcription Factor Assay Kit 10007889

Stability: ≥6 months at -20°C
Summary: Cayman's NF-κB (p65) Transcription Factor Assay detects human NF-κB (p65). It will not cross-react with NF-κB (p50).

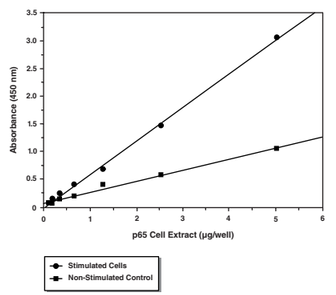
96 wells



NF-κB (human p50/p65) Combo Transcription Factor Assay Kit 10011223

Stability: ≥6 months at -80°C
Summary: Cayman's NF-κB (human p50/p65) Combo Transcription Factor Assay is a non-radioactive, sensitive method for detecting p50 and p65 transcription factor DNA binding activity in nuclear extracts.

96 wells



Nuclear Extraction Kit 10009277

Stability: ≥6 months at -20°C
Summary: Preparation of nuclear extracts is the first step in examining transcription factor activity. Cayman's Nuclear Extraction is formulated for the quick and

simple isolation of nuclear and cytoplasmic fractions from cultured cells and tissue homogenates that can be used successfully in Cayman's Transcription Factor Assay kits. The proteins isolated using this kit can also be used in electrophoretic mobility shift assays (EMSA) and western blotting applications.

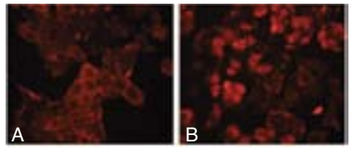
1 ea

p53 Cell-Based Activation/Translocation Assay Kit 600008

Stability: ≥6 months at -20°C

Summary: The tumor suppressor protein p53 plays a crucial role in coordinating cellular responses to genotoxic stress and holds many important clinical implications in the treatment of cancer. Cayman's p53 Cell-Based Activation/Translocation Assay provides a highly specific p53 primary monoclonal antibody together with a DyLight™ (product of Thermo Scientific) conjugated secondary antibody in a ready-to-use format. (-)-Nutlin-3, a potent inhibitor of Mdm2-p53 interaction is included as a positive control.

96 wells



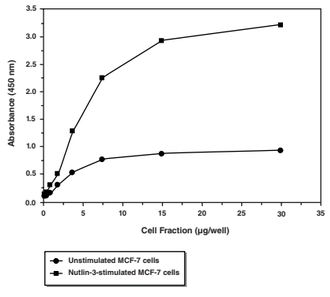
(-)-Nutlin-3-induced translocation of p53 in MCF-7 cells. MCF-7 cells were treated with vehicle *(Panel A)* or 50 µM (-)-Nutlin-2 *(Panel B)* for four hours then fixed and stained with the p53 monoclonal antibody.

p53 Designer Transcription Factor Assay Kit 600030

Stability: ≥1 year at -80°C

Summary: Cayman's p53 Designer Transcription Factor Assay is designed to study alternate p53 DNA-binding sites. A biotinylated oligonucleotide is incubated with p53 contained in a nuclear extract; this mixture then binds to the streptavidin plate provided in the kit. p53 is detected by addition of a specific primary antibody directed against p53. A secondary antibody conjugated to HRP is added to provide a sensitive colorimetric readout at 450 nm.

96 wells

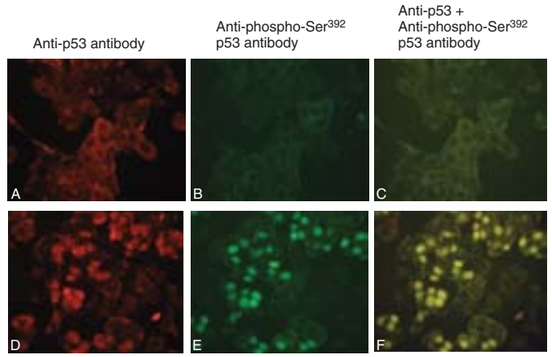


p53 Total and p53 (Phospho-Ser^{392}) Dual Staining Assay Kit 600060

Stability: ≥6 months at -20°C

Summary: Cayman's p53 Total and p53 (Phospho-Ser^{392}) Dual Staining Assay provides a pair of highly specific antibodies against total and phospho-p53 (Phospho-Ser^{392}) together with a pair of matched DyLight™ (product of Thermo Scientific) conjugated secondary antibodies in a ready-to-use format. (-)-Nutlin-3, a potent inhibitor of Mdm2-p53 interaction is included as a positive control.

96 wells



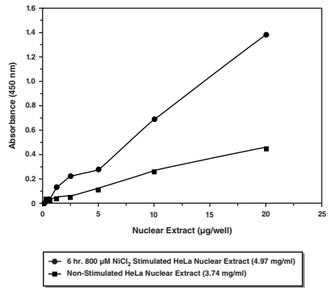
(-)-Nutlin-3-induced translocation of p53 in MCF-7 cells. MCF-7 cells were treated with vehicle *(Panels A-C)* or 50 µM (-)-Nutlin-3 *(Panels D-F)* for four hours then fixed and stained.

p53 Transcription Factor Assay Kit 600020

Stability: ≥1 year at -80°C

Summary: Cayman's p53 Transcription Factor Assay is a non-radioactive, sensitive method for detecting specific transcription factor DNA binding activity in nuclear extracts. A specific dsDNA sequence containing the p53 response element is immobilized onto the wells of a 96-well plate. p53 contained in a nuclear extract binds specifically to the p53 response element and is detected by addition of a specific primary antibody directed against p53. A secondary antibody conjugated to HRP provides a sensitive colorimetric readout at 450 nm.

96 wells



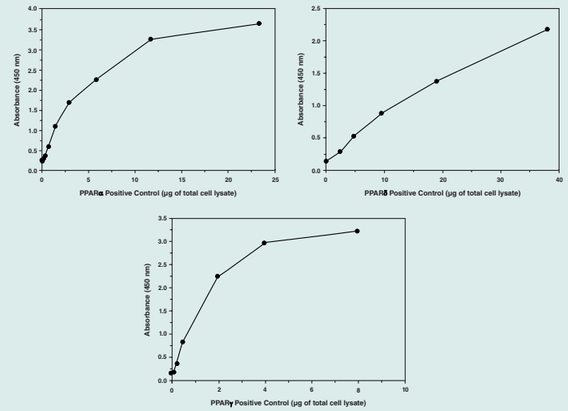
PPAR Transcription Factor Assay Kits

PPARs are ligand-activated transcription factors belonging to the large superfamily of nuclear receptors. PPARα primarily activates genes encoding proteins involved in fatty acid oxidation, while PPARγ primarily activates genes directly involved in lipogenic pathways and insulin signalling. Members of the PPAR family are important direct targets of many antidiabetic and hypolipidemic drugs. Cayman's PPAR Transcription Factor Assays are a non-radioactive, sensitive method for detecting specific transcription factor DNA binding activity in nuclear extracts and whole cell lysates.

PPARα, δ, γ Complete Transcription Factor Assay Kit 10008878

Stability: ≥1 year at -20°C

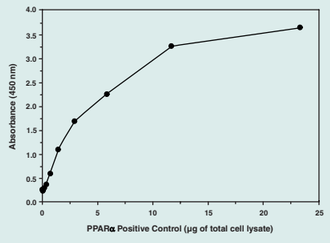
96 wells



PPARα Transcription Factor Assay Kit 10006915

Stability: ≥6 months at -20°C

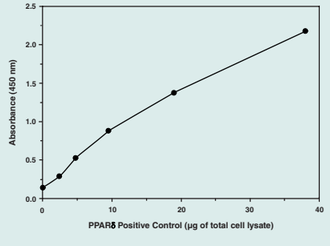
96 wells



PPARδ Transcription Factor Assay Kit 10006914

Stability: ≥6 months at -20°C

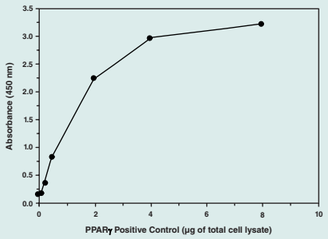
96 wells



PPARγ Transcription Factor Assay Kit 10006855

Stability: ≥6 months at -20°C

96 wells



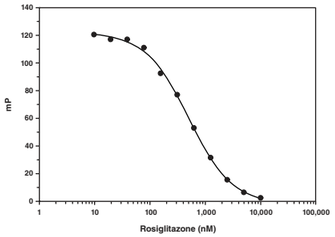
PPARγ FP-Based Ligand Screening Assay Kit - Green 10007685

Stability: ≥6 months at -20°C **Z' Factor:** 0.81

Summary: Cayman's PPARγ FP-Based Ligand Screening Assay - Green provides a fluorescence polarization (FP)-based, single step assay for screening PPARγ ligands. In this assay, a ligand of PPARγ was conjugated to FITC and is used as the displacement probe. Agonists and antagonists of PPARγ will displace the fluorescent probe leading to a decrease in FP. The assay has been validated using known agonists/ligands of PPARγ.

384 wells

1,920 wells

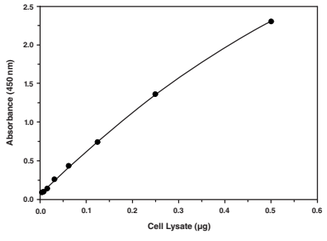


SREBP-1 Transcription Factor Assay Kit 10010854

Stability: ≥1 year at -80°C

Summary: SREBP-1c acts primarily to activate genes required for fatty acid synthesis, such as acetyl CoA carboxylase, fatty acid synthase, and long chain fatty acid elongase. SREBP-1c has important clinical implications in the treatment of many diseases including obesity, diabetes mellitus, insulin resistance, and non-alcoholic fatty liver disease.

96 wells

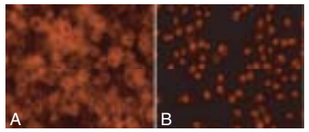


SREBP-2 Cell-Based Translocation Assay Kit 10009239

Stability: ≥1 year at -20°C

Summary: Cayman's SREBP-2 Cell-Based Translocation Assay provides the tools needed to study SREBP-2 movement within whole cells. The kit contains a highly specific SREBP-2 primary antibody together with a DyLight™ (product of Thermo Scientific Inc.) conjugated secondary antibody in a ready to use format. Also included as a positive control is a cholesterol trafficking inhibitor, U18666A.

96 wells

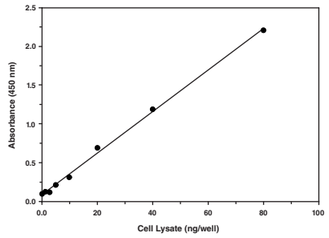


Translocation of SREBP-2 into nuclei by U18666A. RAW 264.7 cells were treated with DMSO (*vehicle; Panel A*) or 24 µM U18666A (*Panel B*) for 72 hours.

SREBP-2 Transcription Factor Assay Kit 10007819

Stability: ≥6 months at -20°C
Summary: SREBP-2 is a transcription factor that performs a critical role in the transcriptional regulation of genes involved in cholesterol synthesis and uptake including HMG-CoA synthase, HMG-CoA reductase, and the LDL receptor.

96 wells



Alphabetical Index

Catalog Number Index