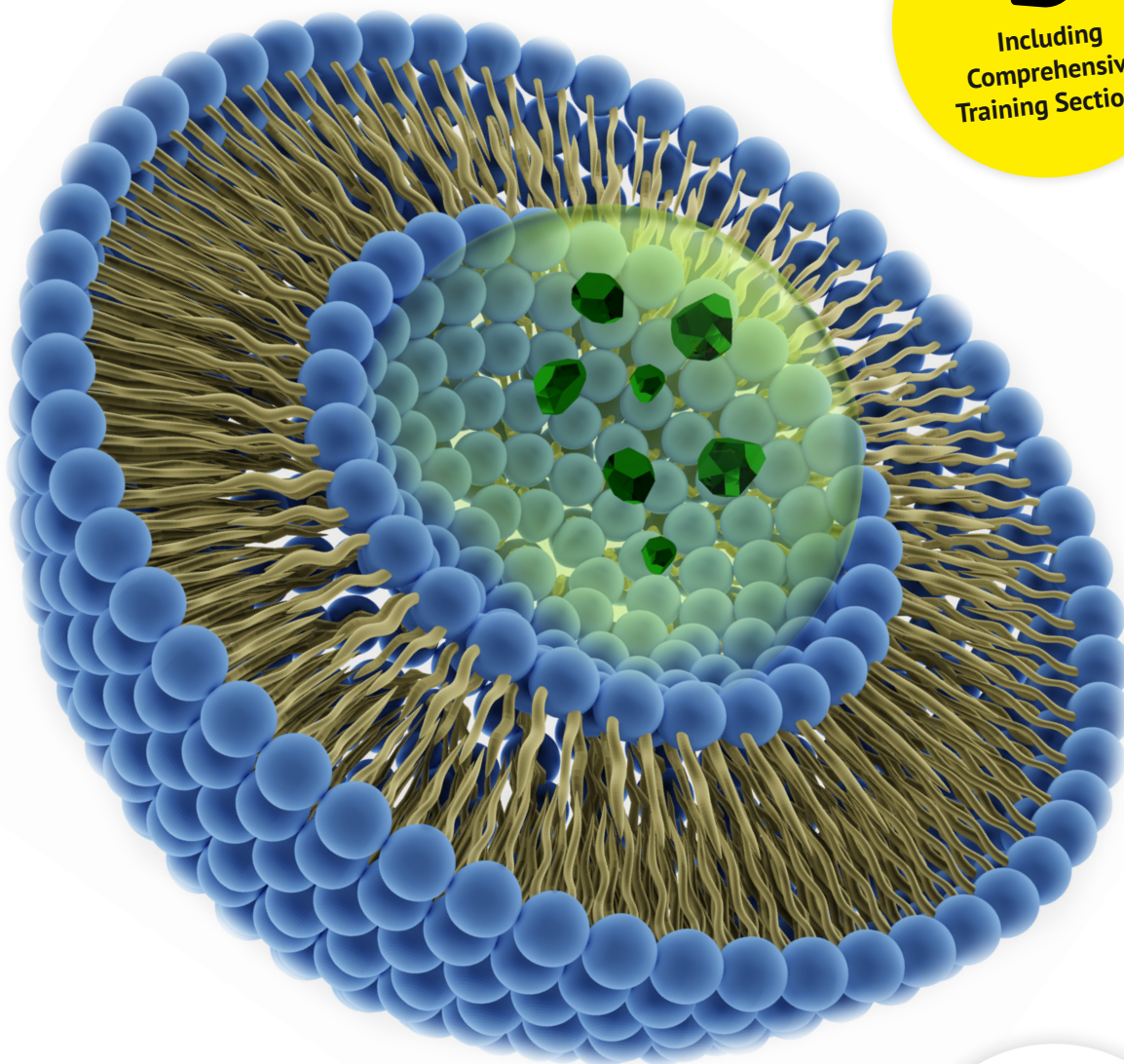




LIPIDOMICS



Including
Comprehensive
Training Sections!



- ▶ Assays and Reagents to Study Lipid Signaling Pathways
- ▶ For Basic Research and Drug Development
- ▶ Manufactured by Echelon Biosciences – The Leader in Lipid Research

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TOPICS IN THIS ISSUE

- ▶ Phosphoinositides – Simplify your PIP Analysis

- ▶ Lipid-Protein Interaction (Part I) – Know your Connections

- ▶ Lipid-Protein Interaction (Part II) – Need Something a Little Different?

- ▶ Lipids – Know your Lipids, Trust your Source

- ▶ Sphingolipids – Echelon’s Expertise, your Science

- ▶ ATX-LPA Made Simple

- ▶ Hyaluronic Acid and Hyaluronidase Assays

- ▶ Antibodies to Phosphoinositides & other Key Lipids

- ▶ Featured Assay – Drug-Induced Phospholipidosis

- ▶ MEP Antibacterial Research

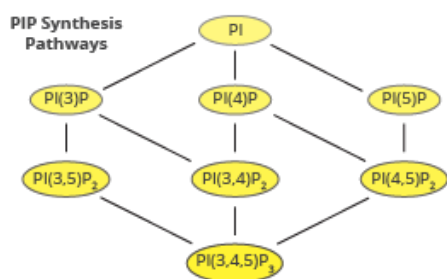
- ▶ Ancillary Products:
 - > Medical-Grade Hyaluronic Acid
 - > Hyaluronic Acid-Sizing Made Easy

Phosphoinositides Quantified

Phosphoinositides – Simplify your PIP Analysis

Phosphatidylinositol phosphates (PIPs) are more than just membrane components. They are vital signaling molecules within the cell, participating in a variety of biological processes ranging from membrane trafficking, inflammation, and cell death.

Echelon Biosciences offers a comprehensive selection of ELISA-based assays to quantify specific PIP levels in cells or tissue samples.



PIP3 Mass Assay – K-2500s

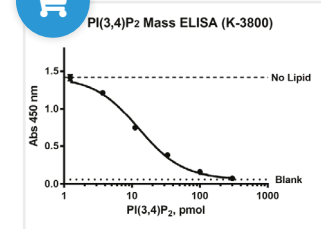
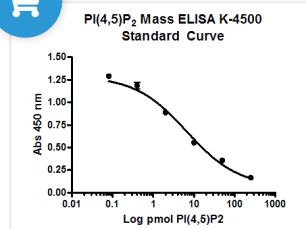
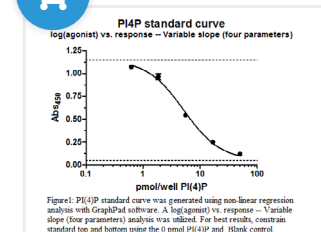
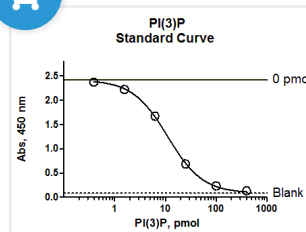
The assay plate method allows the user to determine PI3-K activity by measuring the amount of PIP3 extracted from cells by means of standard ELISA format, eliminating the need for radioactivity and thin layer chromatography.

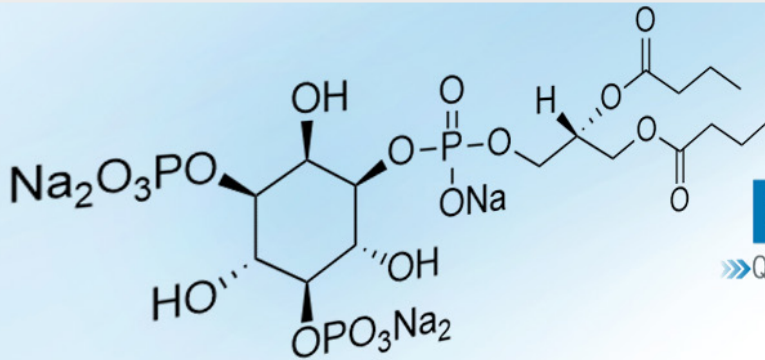


Additionally Featured PIP Mass Assays

Benefits

- ▶ Built-in standards for quantification
- ▶ Analyze 24-32 samples in duplicate or triplicate
- ▶ Assay times are < 4-5 hrs
- ▶ Adaptable to multiple cell types
- ▶ For more information click corresponding pictures

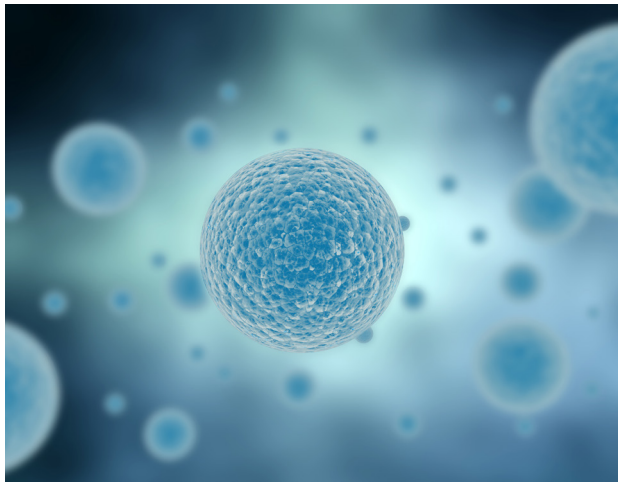




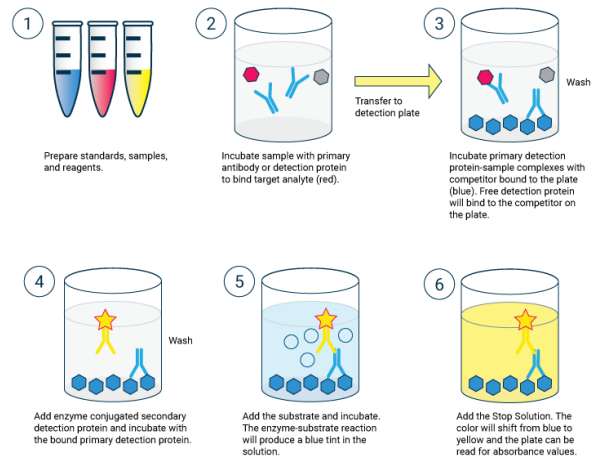
PIP Mass ELISA

Quantify Specific Phosphoinositides in Samples

New to ELISAs?



Running an ELISA is fairly straightforward, but analyzing and interpreting the results can be tricky. Echelon explains one of the most commonly used formats, the Competitive ELISA, and how to interpret the data.



Echelon's ELISAs are centered around biologically interesting lipids and extracellular matrix molecules such as Hyaluronic Acid. Lipids are known to be difficult to use as they are hard to solubilize and tend to stick to tubes and vials. In addition, development of ELISAs generally require a considerable amount of time to maximize the specificity and sensitivity of a binding protein. Detection of the ligand is also complicated by the sample source. Each type of sample brings its own mix of possible cross reactants or interference into the assay.



What is a Competitive ELISA?

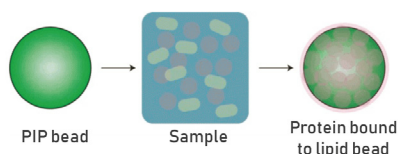
Are your proteins connected to lipids?



Lipid-Protein Interaction (Part I) – Know your Connections!

Lipids are fundamental building blocks of biological membranes and serve as intracellular signaling molecules. An equally important feature of lipids is their influence on protein structure and function. Echelon Biosciences has developed a wide range of reagents for investigating lipid-protein interactions. Their Lipid Coated Beads, FlowPIPs, and PolyPIPosomes can be used in various methods from mass spec to assaying apoptosis.

Lipid Beads



- ▶ For protein pull-down experiments to identify and characterize lipid binding proteins
- ▶ The beads allow lipid binding proteins to be isolated from cell lysates or mixtures of in vitro translated peptides, which can then be analyzed by SDS-PAGE, Western blot, or mass spectrometry

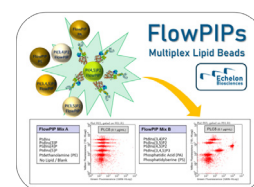
Learn more about experimental applications of Lipid Beads:



Lipid Microparticles: Phagocytosis, Apoptosis, and Beyond

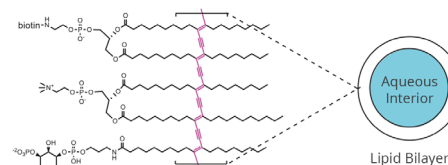
FlowPIPs

- ▶ Multiplex Lipid Beads are microparticles with attached lipids
- ▶ Excellent choice to study lipid binding proteins in a multiplexed flow cytometry application

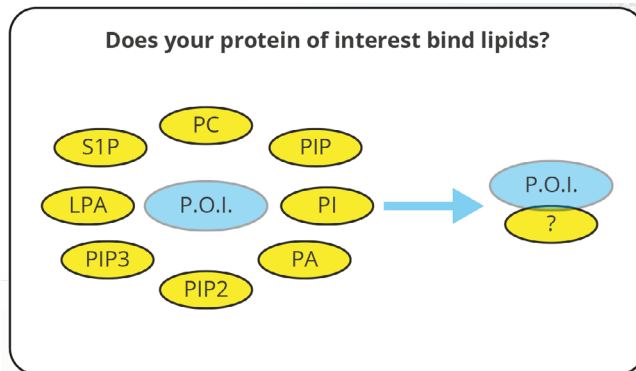


PolyPIPosomes

- ▶ Ready-to-use polymerized liposomes composed of specific phosphoinositides



- ▶ Ideal for numerous assays, including: protein pull-down, surface plasmon resonance, and liposome overlay assay
- ▶ Markedly increased stability (up to 6 months) vs. conventional liposomes
- ▶ A biotin tag is incorporated for easy detection of PIPn-binding proteins with streptavidin reagents



Lipid-Protein Interaction (Part II) – Need Something a Little Different?

Lipids bind and influence the structure and function of proteins. While these interactions are critical, studying them can be challenging. Echelon Biosciences has developed a set of Lipid Strips that lower the barriers for interrogating lipid-protein binding partners. Their PIP Strips, Membrane Strips, and Sphingo Strips provide a simple and easy method for determining novel lipid-protein interactions with purified proteins.

PIP Strips (P-6001)

PIP Strips are 2 cm x 6 cm hydrophobic membranes that have been spotted with 100 pmol of all eight phosphoinositides and seven other biologically important lipids. These strips can be used to determine if a protein of interest interacts with one or more of these lipids in a simple protein-lipid overlay assay.

Lysophosphatidic Acid	○	Sphingosine-1-phosphate
Lysophosphocholine	○	PtdIns(3,4)P2
PtdIns	○	PtdIns(3,5)P2
PtdIns(3)P	○	PtdIns(4,5)P2
PtdIns(4)P	○	PtdIns(3,4,5)P3
PtdIns(5)P	○	Phosphatidic Acid
Phosphatidylethanolamine	○	Phosphatidylserine
Phosphatidylcholine	○	Blue Blank

Benefits

- ▶ Simultaneous screening of multiple lipids
- ▶ The procedure is very similar to a Western blot. The membrane is blocked, incubated with a lipid binding protein of interest followed by primary and secondary antibodies. The PIP Strips

are then visualized with chemiluminescence, fluorescence, or with precipitating TMB.

- ▶ From Science to Nature to Cell, there are hundreds of publications citing PIP Strips from Echelon Biosciences



Membrane Strips (P-6002)

- ▶ A nitrocellulose membrane with 15 different biologically important lipids found in cell membranes



Sphingo Strips (S-6000)

- ▶ A nitrocellulose membrane spotted with 15 different sphingolipids



Do you know what lipid your protein binds to but need to know how well? No problem!

Lipid Arrays (P-6100 , P-6003 , and S-6001) use a concentration gradient of lipids to determine the degree of binding of your protein.



5 Tips to Improve Your Strips!

Don't wipeout due to impure lipids



Lipids – Know your Lipids, Trust your Source

Synthetic, pure, reliable. Don't accept anything less than the highest quality lipids from Echelon Biosciences. Their scientists have years of experience in lipid biology. Hundreds of different lipids are available.




- Phospholipids
- Phosphoinositides
- Sphingolipids
- Isoprenoids
- Liposome components
- Bioactive lipids
- Deuterated analogs
- And many more

Featured Lipids:




Phosphoinositides & Derivatives

From PIPs with various acyl chain lengths to biotin and fluorophore conjugates, we have your phosphoinositides needs covered.

▶ PI(4,5)P2:





- ▶ Phosphatidylinositol 4,5-bisphosphate diC4 (P-4504) 
- ▶ Phosphatidylinositol 4,5-bisphosphate diC8 (P-4508) 
- ▶ Phosphatidylinositol 4,5-bisphosphate diC16 (P-4516) 

▶ Labeled PIPs:

- ▶ BODIPY FL PI(3)P is a water soluble analog of PI(3)P labeled with a green fluorophore: C-03F6 
- ▶ BODIPY FL Phosphatidylinositol 4,5-bisphosphate is a water soluble analog of PI(4,5)P2 (PIP2) labeled with a green fluorophore: C-45F6 
- ▶ BODIPY FL PI(3,4,5)P3 is a water soluble analog of PI(3,4,5)P3 (PIP3) labeled with a green fluorophore: C-39F6 

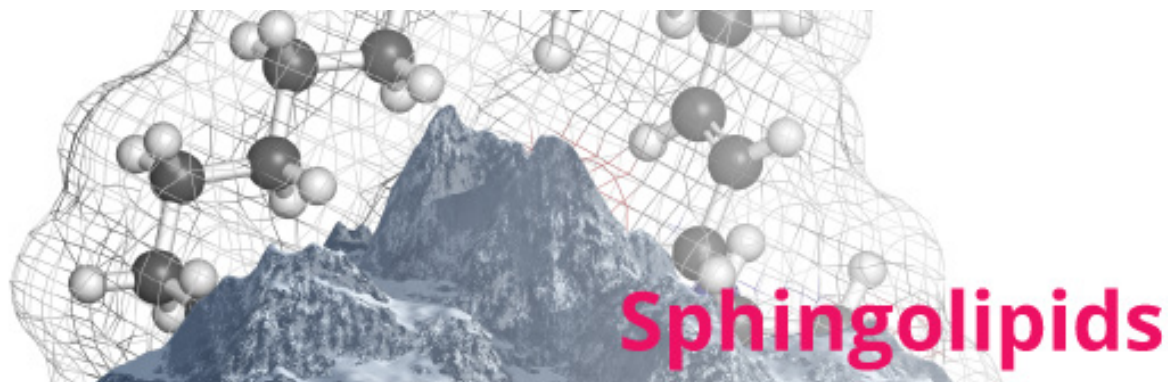
New Glycolipids!

Glycolipids have important roles in cell membrane stability and facilitating immune responses. They are also useful as surfactants and emulsifying agents for a variety of applications.

- ▶ Decyl rhamnoside: RL10-1 
- ▶ L-Ascorbyl-6-stearate: R-1218 
- ▶ Rha(rac)C10C10: RL10105 
- ▶ Rha(rac)C12C12: RL12125 



Isoprenoids: Biology & Potential Uses







Sphingolipids – Echelon’s Expertise, your Science

Sphingolipid biology includes membrane structures, inflammation, immune cell activation, and recognition of exogenous agents. Sphingolipids are built around a sphingosine backbone via de-novo synthesis in the ER from fatty acid-CoA and serine. Defects in sphingolipid metabolism cause many human diseases.

Sphingolipid Detection

From ELISAs to antibodies, we’ve got you covered!

- ▶ Sphingosine 1-Phosphate ELISA: K-1900 
- ▶ Sphingosine Kinase Activity Assay: K-3500 
- ▶ Neutral Sphingomyelinase Activity Assay: K-1800 
- ▶ S1P Antibody: Z-P300 

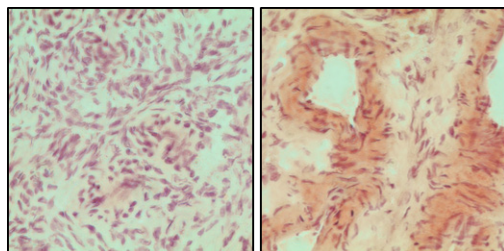





Figure: Immunohistochemistry with S1P Antibody in Red.
Sphingosine 1-Phosphate (S1P) is a key component of the sphingolipid signaling cascade. Recent research has suggested that S1P is a potent tumorigenic growth factor and may be a novel biomarker for early stage cancer detection.

Sphingolipid-Protein Interaction Tools

Understanding lipids can be difficult, but the experiments don’t have to be. Echelon Biosciences provides a variety of tools to uncover sphingolipid-protein interactions.





Featured Products

- ▶ Sphingolipid Coated Beads 
- ▶ Binding Strips and Arrays:
 - S-6001 
 - S-6000 

Sphingolipid Biochemical Reagents

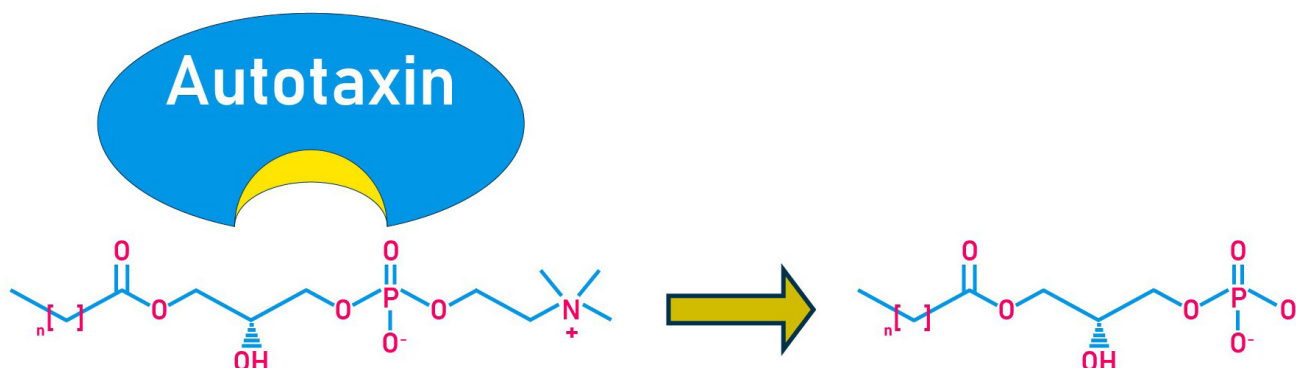
An extensive list of analogs, inhibitors, enzymes, and receptor antagonists.

Featured Products

- ▶ S1P Receptor Antagonist (B-0061) 
- ▶ Sphingosine Kinase 1 (E-K068) 
- ▶ Ceramide 1-Phosphate (S-5018) 
- ▶ Biotin Sphingomyelin (S-400B) 



Sphingolipid Function in Biology






ATX-LPA Made Simple

The Autotaxin-Lysophosphatidic Acid (ATX-LPA) axis is a well-characterized signaling pathway that is known to be aberrantly regulated in many types of cancer. To date, ATX remains an attractive target for drug discovery due to its role in cancer stem cells and the tumor microenvironment.




Echelon Biosciences' line of ATX and LPA assays and reagents has everything you need to move your research forward.

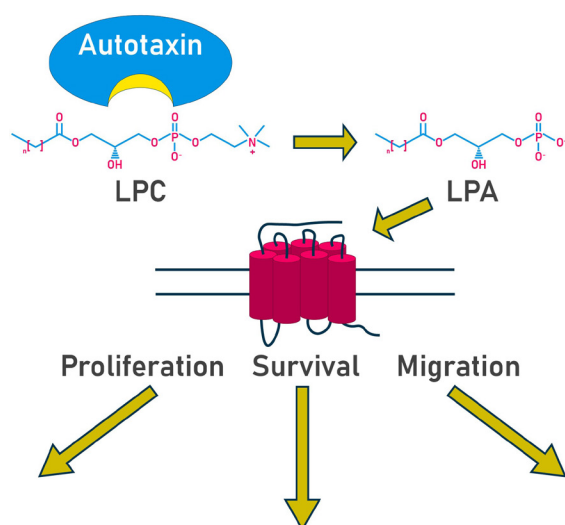
Autotaxin

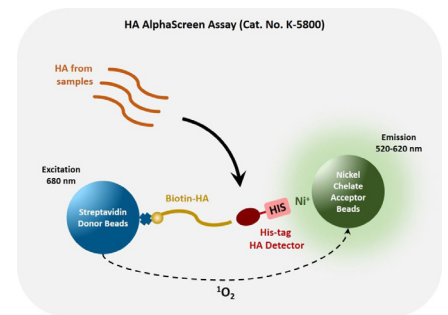
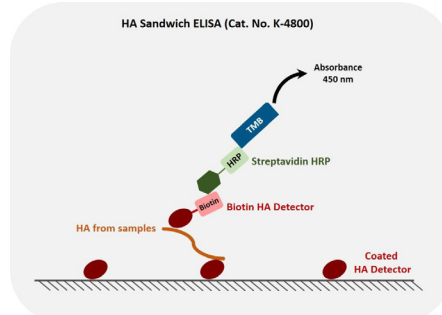
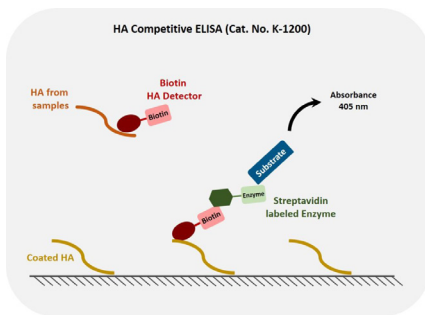
Featured Products:

- ▶ Autotaxin Inhibitor Screening Kit: K-4200 
- ▶ ATX Antibody: Z-P400 
- ▶ ATX Enzyme, active: E-4000 

Lysophosphatidic Acid

- ▶ LPA ELISA: K-2800S 
- ▶ LPA Antibody (504B3): Z-P200 
- ▶ LPA - Lipids and Conjugates 





Hyaluronic Acid and Hyaluronidase Assays

Hyaluronic Acid Quantification

Next to its huge portfolio for Lipid Research, Echelon Biosciences offers a unique Hyaluronic Acid (HA) product line. HA, also called hyaluronan, is a glycosaminoglycan distributed widely throughout connective, epithelial, and neural tissues. The molecule is also present in human blood samples and synovial fluids. As one of the main components of the extracellular matrix, HA contributes significantly to cell adhesion, cell proliferation, and migration.

HA Assays

Echelon Biosciences offers 3 different assays to detect HA from biological samples.

- ▶ Hyaluronan Enzyme-Linked Immunosorbent Assay: K-1200
- ▶ Hyaluronic Acid Sandwich ELISA: K-4800
- ▶ Hyaluronic Acid AlphaScreen Assay: K-5800



All are high-quality assays with low assay coefficient of variation (CV), sensitive detection, and easy-to-use designs. The graphic above provides illustrations of the formats.



Which Hyaluronic Acid Assay Should I Use?

Hyaluronidase Assays

Hyaluronidases are a group of enzymes that degrade HA and are involved in fertilization and wound healing and in several pathological processes. The Hyaluronidase Activity ELISA is a quantitative immunoassay designed for the in vitro measurement of hyaluronidase activity in biological samples.

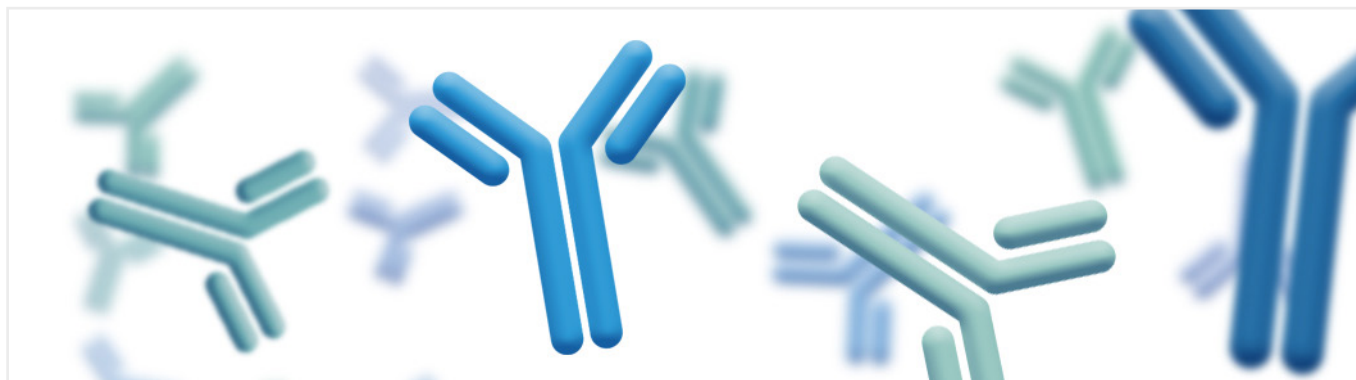
- ▶ Hyaluronidase Activity ELISA: K-6000



Learn more about the potential connection between Hyaluronidase, HA, and COVID-19:



Hyaluronidase & COVID-19



Antibodies to Phosphoinositides & other Key Lipids

Lipid biology requires appropriate tools for visualizing and understanding their function. Echelon Biosciences carries an array of lipid antibodies that have been validated in a range of applications, including ICC and IHC.

PIP Antibodies

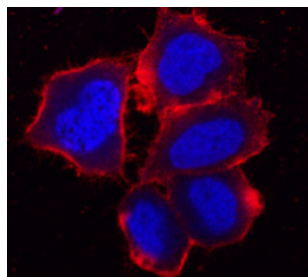


Figure (left):
Immunocytochemistry with
Anti-PI(4)P (Z-P004) in Red.
Phosphatidylinositol 4-phosphate
(PI(4)P) is the biosynthetic precursor
to PI(4,5)P2 and has an important
role in regulating sphingomyelin and
glycosphingolipid metabolism and
membrane trafficking at the exit of the
Golgi complex.

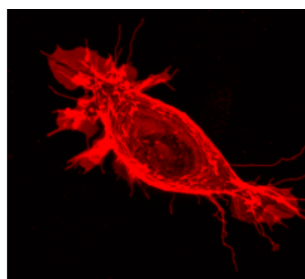
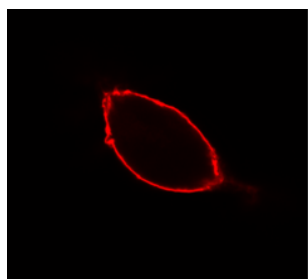


Figure (above):
Mouse monoclonal antibody targeting PI(4,5)P2,
Immunofluorescence (Z-P045)



LPA Antibody

Lysophosphatidic Acid (LPA) is a serum-derived phospholipid involved in diverse cellular processes. Recent research indicates LPA may play a significant role in the pathophysiology of cancer and may be used as a biomarker for ovarian cancer.

The LPA antibody (Z-P200) is validated for use in IHC, ICC, and ELISA.



DGK Antibodies

Echelon Biosciences' anti-DGK α (Z-DGKA) and anti-DGK ζ (Z-DGKZ) antibodies recognize specific isoforms of diacylglycerol kinase (DGK). DGK catalyzes a reaction terminating the protein kinase C-mediated signaling by converting diacylglycerol (DAG) to phosphatidic acid (PA), a signaling lipid which is involved in DNA synthesis, phospholipid synthesis, and regulation of several enzymes.

▶ Anti-DGK α antibodies (Z-DGKA)



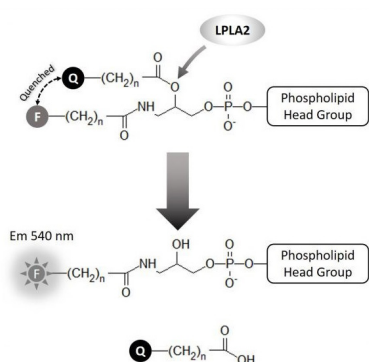
▶ Anti-DGK ζ antibodies (Z-DGKZ)



Lipid Immunostaining



Featured Assay – Drug-Induced Phospholipidosis



Lysosomal PLA2 Inhibitor Screen

Identify problematic compounds early in the drug discovery process!

The FDA has determined drug-induced phospholipidosis (DIPL) is a serious drug safety issue. Evidence is accumulating that DIPL is the result of certain cationic amphiphilic drugs (CADs) directly inhibiting Lysosomal Phospholipase A2 (LPLA2).

Echelon Biosciences' LPLA2 Inhibitor Screen (K-7000I) is designed to assay a drug's ability to inhibit LPLA2 activity in vitro. The LPLA2 inhibitor screen could predict if a drug will induce phospholipidosis.



SLAS-Poster:

A Reliable High-Throughput Enzymatic Screen for Drug-Induced Phospholipidosis

Open Poster

LPLA2 Related Products

- LPLA2 Activity Assay (K-7000A) :
A homogenous assay designed to detect LPLA2 activity from both biological and purified sources of LPLA2
- Human Lysosomal Phospholipase A2 (hLPLA2), active (E-7000):
Recombinant, full-length human, lysosomal phospholipase A2 (LPLA2) protein



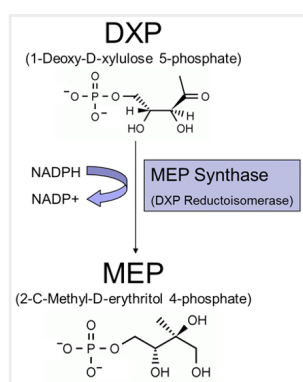
Drug-Induced Phospholipidosis: Best Practices and Screening



MEP Antibacterial Research

Echelon Biosciences is the only source for reagents for every step of the Methylerythritol Pathway (MEP), including intermediates, assays, and testing services.


The MEP pathway is used by most bacteria, including all Gram-negative bacteria, for isoprenoid biosynthesis. Isoprenoids comprise one of the most diverse classes of compounds found in nature. With over 50,000 different isoprenoids identified to date, they exhibit a broad range of structural complexity and are involved in a variety of biological functions including electron transport (quinones), stabilization of cell membranes (hopanoids and sterols), cell wall biosynthesis (dolichols), signal transduction (prenylated proteins), photosynthesis (chlorophylls), and modification of tRNAs.





Two unrelated essential pathways exist in nature for the biosynthesis of isoprenoid metabolites, which include the MEP pathway, unique to bacteria, and the mevalonate (MVA) pathway used by humans.

The individual enzymatic steps of the MEP pathway are attractive for the development of new antibiotics.

Featured Products:

- ▶ **MEP Synthase (DXR) Enzyme Inhibitor Screen (K-2000C)** 

The assay will evaluate compounds for inhibition of DXR activity. It is a valuable tool for the identification of potential antimicrobials.
- ▶ **MEP Synthase (DXR), active (E-2000C)** 

Recombinant C-terminal His-tagged, MEP Synthase
- ▶ **2-C-Methyl-D-erythritol 4-phosphate (MEP) (I-M051)** 

Isoprenoid; provided as sodium salt

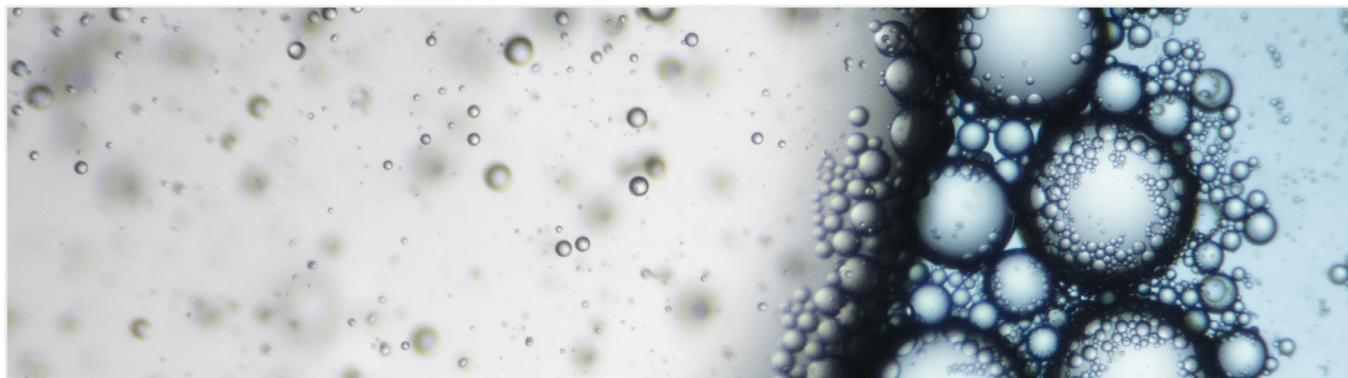
SLAS-Poster:

Drug Targeting of Novel Antimicrobials in the MEP Pathway

 **Open Poster**

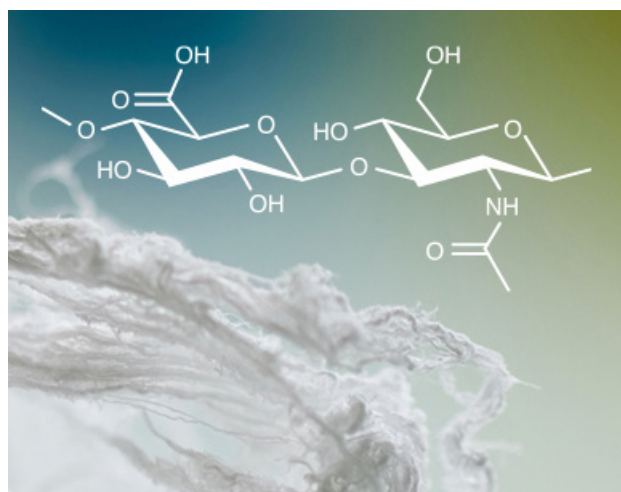


MEP Antibacterial Research



Ancillary Products

Echelon Biosciences offers the following products in close cooperation with highly recognized companies.



Medical-Grade Hyaluronic Acid (HA):

- ▶ High-quality HA: medical-grade, low impurity contents
- ▶ Fast dissolving and easy to handle HA
- ▶ Batch-to-batch consistency ensuring reproducibility of experiments and easy scale-up
- ▶ Certificates of Analysis and polydispersity index information are available for every batch
- ▶ Various molecular weights of HA available

Hyaluronic Acid-Sizing Made Easy

Hyaluronic acid (HA) is an oft-studied glycosaminoglycan found in connective, epithelial, and nervous tissues. Tissue HA is naturally turned over each day with a fraction released to circulate in biological fluids. Elevated serum HA concentration has long been used as a biomarker for liver disease. Recent work established that not only the amount but also the size of HA is altered in disease-related pathological states. For example, low molecular weight serum HA can be used to differentiate metastatic from non-metastatic breast cancer.

Traditional HA sizing was done by expensive and time-consuming size exclusion chromatography with a multi-angle laser light scattering detector. Echelon now offers Hyalose Select-HA™ Ladders providing an easy and affordable way to analyze HA size using a common laboratory technique – agarose gel electrophoresis.



**Select-HA™ –
For Tight Size Control: Product Overview**



**How to Determine
Hyaluronic Acid Molecular Weight
Using Gel Electrophoresis**



Innovative Tools for Molecular and Cell Biology

MoBiTec GmbH

MoBiTec develops cutting-edge research tools for the global life science, diagnostic, and pharmaceutical community. These include versatile purification columns and unique vector systems for protein expression in different species. Moreover, MoBiTec is specialized in labeling and detection technologies. Our products are distributed worldwide.

Next to our own portfolio, MoBiTec is a leading distributor for innovative life science reagents and services. We represent a number of well-established international companies:



For the most up-to-date listing of our supplier companies please visit our website

www.mobitec.com



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