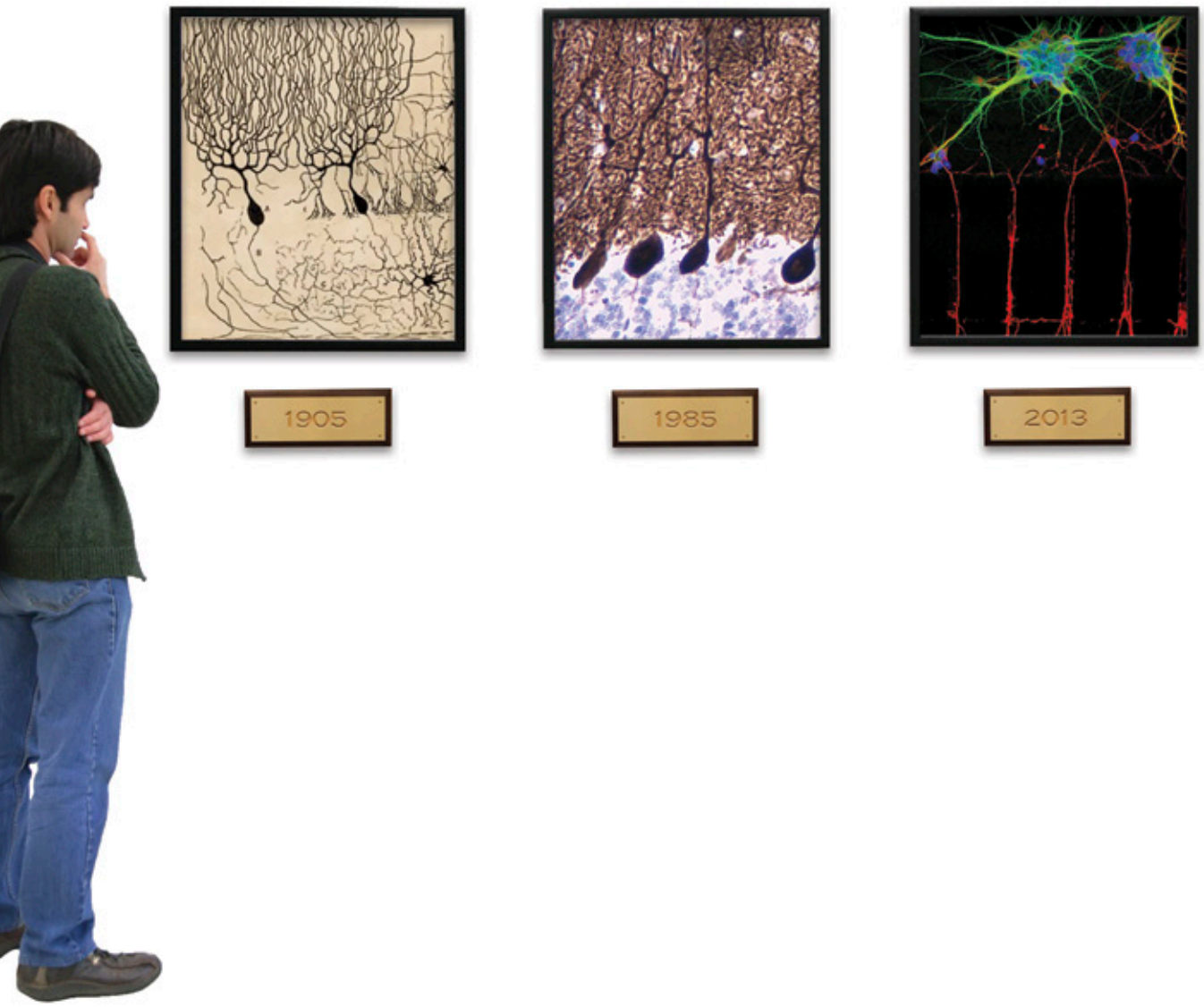
The Art of Neuroscience

Antibodies, proteins, kits, and assays



Merck Millipore is a division of MERCK

Platforms, Technologies, and Services

As a tools provider and partner in research, Merck Millipore is committed to the advancement of life science research and therapeutic development. This guide includes a number of new products for target identification, pathway detection, and profiling. These products provide proven solutions for a range of applications and are backed by extensive technical support.

STEM CELLS AND PRIMARY CELLS

Merck Millipore offers an extensive range of embryonic, neural, and mesenchymal stem cells for both human and rodent studies. This includes novel human neural stem cells, human embryonic stem cells, and a complete line of mouse embryonic stem cells. Endothelial and epithelial cells are also available.

CELL CULTURE SYSTEMS

Merck Millipore's innovative cell culture solutions help optimize cell growth and maintenance for neuroscience research. We offer media designed for many cell types, including serum-free, feeder-free formulations specifically validated for neural stem cells. Our flexible sterile filtration devices offer fast flow and have many membrane options. Also available are microfluidic perfusion systems and cultureware to mimic *in vivo* conditions and enable dynamic live cell analysis.

ANTIBODIES AND IMMUNOASSAYS

Merck Millipore's highly validated antibodies are guaranteed for quality performance. In addition to the validation, all our antibodies are backed with a best-in-industry technical service team dedicated to our antibodies customers. With the combined expertise of Chemicon® and Upstate®, we offer the most comprehensive portfolio of antibodies and assays across the life sciences industry. For precise quantification of soluble targets from sera and lysates, and Merck Millipore provides a growing number of ELISAs for inflammation, neurotrophins, and neurodegenerative disease biomarkers.

CELL-BASED ASSAYS AND QUANTITATIVE CELL IMAGING

Merck Millipore offers a significant portfolio of live cell, whole-cell and cell-based activity assays and reporter systems for direct and indirect detection. These technologies facilitate protein target validation, identify cellular pathways and determine mechanism of action for lead optimization environments.

Cover:

1905: Ramon y Cajal Golgi staining of cerebellar cell types

1985: IHC staining of human cerebellum using anti-INSP3R (Catalogue No. ABS55) and HRP/DAB conjugated secondary.

2013: Confocal imaging of triple fluorescent IHC staining of cultured neurons in microfluidic axon investigation chamber (AXIS®) (Catalogue No. AX15005PBC). Somas and dendrites are shown (green) stained with anti-MAP2 (Catalogue No. MAB3418) and Alexa Fluor® 488. Axonal staining (red) is shown using anti-βIII tubulin (Catalogue No. AB15708) and CY3. Nuclei are counterstained (blue) with DAPI.

FLOW CYTOMETRY ASSAYS AND SYSTEMS

Simultaneously measuring multiple parameters on individual cells, flow cytometry is essential for in-depth cell analysis. Our Amnis® imaging flow cytometers combine the speed, sensitivity, and phenotyping abilities of flow cytometry with the imagery and functional insights of microscopy, taking neuroscience to higher levels of discrimination and discovery. Our easyCyte™ flow cytometers provide precise measurement via microcapillary technology that translates into smaller samples, less reagents, and minimal waste. Validated FlowCellect® assay kits, Milli-Mark™ conjugated antibodies and application-specific software modules provide a complete solution for flow cytometry.

MILLIPLEX® MAP MULTIPLEX ASSAYS

MILLIPLEX® MAP assays offer the broadest selection of multiplex kits and reagents in a wide variety of research areas, including stem cell differentiation, inflammation, metabolism, and neurodegeneration. MILLIPLEX® MAP enables the simultaneous detection of multiple soluble or intracellular biomarkers using a small sample size. Using the Luminex® xMAP® bead-based technology, Merck Millipore's flexible and customizable assays are exhaustively tested and qualified for sensitivity, specificity, reproducibility and wide dynamic range.

CALBIOCHEM® COMPOUNDS

Merck Millipore's Calbiochem® line of high quality inhibitors, biochemicals, antibodies, proteins, and kits have been cited in thousands of peer-reviewed publications. Small-molecule compounds, including inhibitors, activators, and other pathway modulators, are critical tools for researchers studying cell signaling and other intracellular mechanisms that control cell fate, function and phenotype. From libraries and pathway panels to individual reagents, the Calbiochem® line of products offers the widest and most cited selection of inhibitors and activators worldwide.

Introduction

Neuroscience: a synapse of old and new technologies

Understanding our complex brain requires a coordination of several diverse research areas. From biophysics to molecular biology, cell physiology, and cognitive neuroscience, researchers are using a broad range of tools. Though the difficulties in studying neural systems are rooted in their incredibly complex and delicate configurations, many experimental challenges have been surmounted by key research tools that are not quantum-leap advancements but are rather amalgams of old and new technologies.

The synergy of the old and the new is epitomized by modern applications of classic immunochemistry, in which a specific antibody combines with an antigen to form a unique antibody-antigen complex. Using antibodies to probe for individual macromolecular epitopes is particularly tractable because the highly specific antibodies can be created, without laborious engineering, by a host immune response. The principle of antibody-antigen specificity has laid the foundation for both the simplest immunodetection assays as well as complex applications like multiparametric biomarker and whole cell analysis.

Antibody-based biomarker detection reagents are now available for numerous cell and tissue types, disease states, and for measuring responses to targeted therapies. These extremely specific, robust immunoprobes are now used with high-tech fluorescent secondary antibodies and powerful microscopes, keeping alive the classic technique of immunocytochemistry in neuroscience research. Western blotting, another established immunodetection technique for cell and tissue lysates, has been vastly improved by membrane (and probe) technology. From the old technique of immunoprecipitation has evolved chromatin immunoprecipitation (ChIP), in which antibodies recognize specific proteins bound to DNA. The field of epigenetics owes most of its growth to improvements in ChIP. The nascent method, RIP-chip (RNA-binding protein immunoprecipitation coupled with microarray analysis) promises to be equally valuable.

Multiparametric biomarker detection also combines old and new technologies. Advances based on the classic enzyme-linked immunosorbent assay (ELISA) for biomarker quantification have yielded multiplex assays that exploit antibody-fluorophore conjugates for efficient, precise measurement of nearly 100 analytes per sample. Even the technique of flow cytometry, historically limited to users with specialized expertise, has vastly broadened with the availability of precision antibody-based reagents for biomarker detection. New, inexpensive systems and good neuro-specific biomarkers have granted flow cytometry access to neuroscientists in cancer research and regenerative medicine.

Neuroscience is a heterogeneous field, requiring integrated analysis of multiple cell types, tissues and organs, using diverse techniques. With which of these techniques will the next breakthrough be made? Given that researchers tend to build upon traditional technologies rather than abandon them, the next advance in neuroscience will likely rely on antibodies and immunodetection.

Featured Products

- ReNcell® Human Neural Stem Cells PG 6
- Calbiochem® InhibitorSelect™ Protein Kinase Inhibitor Libraries for NSCs PG 7
- Human Oligodendrocyte Differentiation Kit PG 9
- N21 Medium Supplement PG 10
- InhibitorSelect™ Wnt Signaling Pathway Inhibitor Panel PG 14
- InhibitorSelect™ Hh Signaling pathway Modulators PG 15
- Magna ChIP® G Tissue Kit PG 20
- InhibitorSelect™ PI3-K/Akt/mTOR Signaling Pathway Inhibitor Panel PG 30
- MILLIPLEX® MAP Human Neurodegenerative Disease Magnetic Bead Panels PG 37
- LentiBrite™ GFP-LC3 & GFP-LC3 Mutant Lentiviral Biosensors PG 40
- LentiBrite™ PSD95-RFP & PSD95-GFP Lentiviral Biosensors PG 40

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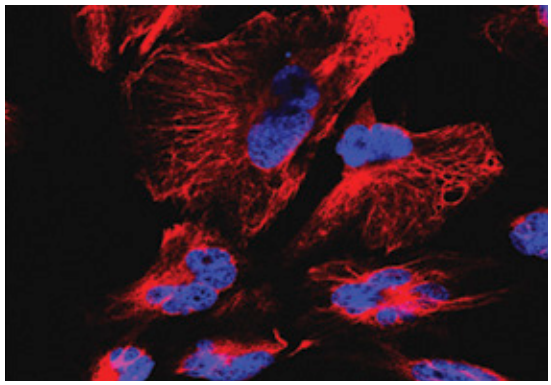
Development

Nervous system complexity requires an intricate, precise developmental plan. Patterns of cell differentiation, signaling, migration, neurite outgrowth, and synapse formation occur across the embryo with unparalleled choreography. The broad array of immunodetection techniques now available and emerging technologies in genetics and epigenetics are helping to reveal the processes underlying brain development and disease.

Neural Stem Cells

The discovery that neurons, astrocytes, and oligodendrocytes arise from neural stem cells (NSCs) has created new opportunities for treating nervous system diseases. Merck Millipore's extensive array of human and rodent neural stem cell systems includes novel cell lines derived from both adult and embryonic neural tissue, optimized media for cell expansion and differentiation, and a complete selection of lineage-specific and stage-specific antibodies.

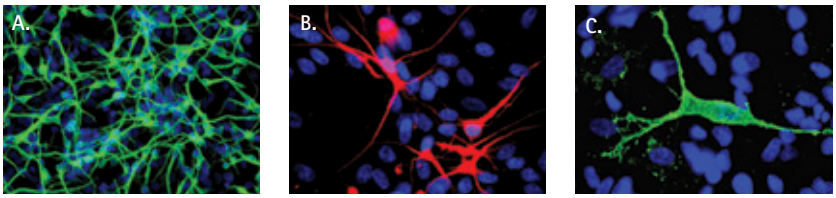


Immunocytochemistry Analysis: Representative lot data. Epifluorescent analysis of human WI38 cell line using Anti-Nestin, human antibody (red). Nuclei are stained with DAPI counterstain (Blue). This antibody positively stains nestin filaments in the human WI38 cell line.

Anti-Nestin

(Catalogue No. ABD69)

Nestin, a large intermediate filament protein (class Type VI), is expressed during development and in myotendinous and neuromuscular junctions. Nestin identifies the primitive neuroepithelium and many other embryonic tissues.



Multipotentiality of ReNcell® cells. Both ReNcell® CX and ReNcell® VM cell lines are readily differentiated into all three neural phenoytpes: neurons (βIII-tubulin, green, 20X, A); astrocytes (GFAP, red, 40X, B) and oligodendrocytes (Gal C, green, 60X, C); all counterstained with Hoechst nuclear stain (blue).

ReNcell® Human Neural Stem Cells

(Catalogue Nos. SCC009 and SCC010)

ReNcell® VM cells and ReNcell® CX cells are well-established NSC lines derived from the ventral mesencephalon and cortex of the developing human brain, respectively, and transfected with myc. Both cell lines offer easy maintentenance, phenotype and genotype stability, and multipotential differentiation capacity over long-term culture. ReNcell® lines differentiate readily in response to the condition of interest, making them the ideal platform for neuroscience research.

Calbiochem® InhibitorSelect™ Protein Kinase Inhibitor Libraries

Two panels of kinase inhibitors, InhibitorSelect™ 96-Well Protein Kinase Inhibitor Library I and InhibitorSelect™ 96-Well Protein Kinase Inhibitor Library II, were screened to identify chemicals that optimize and increase the propagation of Neuroepithelial Stem (NES) cell cultures. These libraries make up a potent, specific, pharmacologically active, well-characterized, and structurally diverse set of 160 compounds.

For a complete table of inhibitor targets included in each panel, visit **www.merckbiosciences.com/inhibitorselect**.

Inhibitor Characteristics

- Cell-permeable†
- Potent and selective†
- ATP-competitive†
- Reversible†
- Stable in DMSO
- Structurally diverse
- Some target multiple kinases
- Known pharmacological activity
- Less toxic

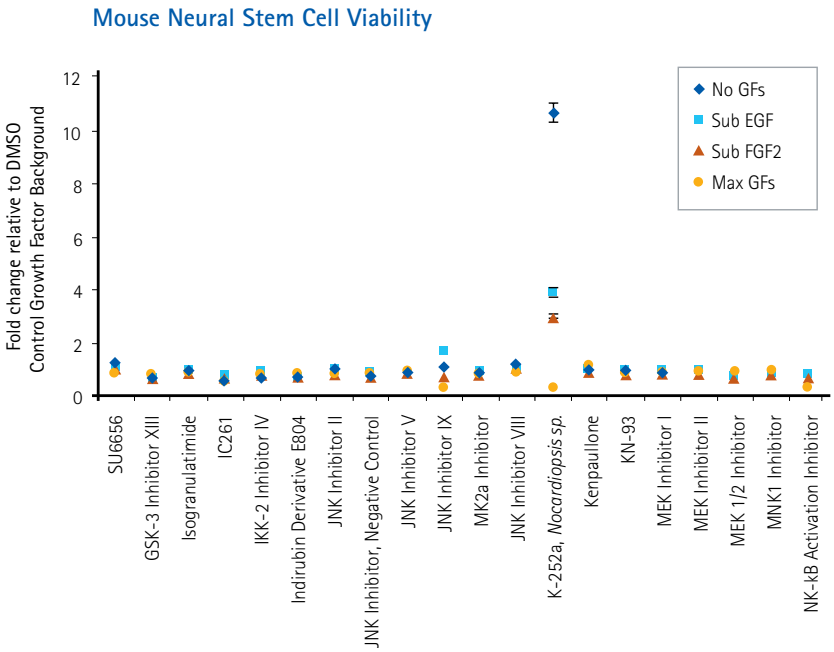
Highest Quality Control

- Purity by HPLC (≥95%)†
- Lot-specific data for every inhibitor in solution
- DMSO-resistant polypropylene deep-well microplates

CD-ROM with comprehensive data set included in each library

- Inhibitor description
- Published IC_{50}/K_i values
- Literature citations
- SD files
- CAS numbers
- PubChem compound ID
- Molecular weight
- Molecular structure

†Pertains to the majority of inhibitors.



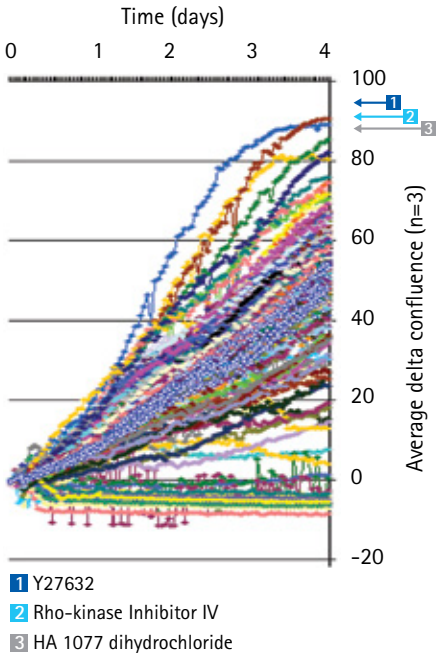
InhibitorSelect™ 96-Well Protein Kinase Inhibitor Library I

(Catalogue No. 539744)

This panel of compounds consists of 80, well-characterized protein kinase inhibitors targeting mainly tyrosine, AGC, and atypical families of kinases, the majority of which are cell-permeable and ATP-competitive. The library is useful for cancer signaling pathway analysis, cell-based assays, target identification in drug discovery, screening new protein kinases, and other related applications. It is supplied with a CD containing comprehensive documentation for each inhibitor.

Graph (left): InhibitorSelect™ 96-Well Protein Kinase Inhibitor Libraries I & II (160 inhibitors; Cat. Nos. 539744 and 539745) were screened for influence on proliferation and survival of mouse neural stem cells (mNS) in a cell viability assay under 4 conditions: (A) No GFs – No Growth Factors (to identify survival/proliferation factors) (B) Sub EGF – Sub-optimal EGF (to identify inhibitors/potentiators) 20 pg/mL EGF (C) Sub FGF2 – Sub-optimal FGF2 (to identify inhibitors/potentiators) 500 pg/mL FGF2 (D) Max GFs – Maximal EGF + FGF2 (to identify inhibitors/potentiators) 20 ng/mL EGF + 20 ng/mL FGF2 The presence of inhibitor K-252a, *Nocardiopsis sp.* (Cat. No. 420297) alone in the culture medium resulted in a 10-fold mNS cell viability.

Data courtesy of Donna McLaren, Stem Cell Sciences, Cambridge, UK



InhibitorSelect™ 96-Well Protein Kinase Inhibitor Library II

(Catalogue No. 539745)

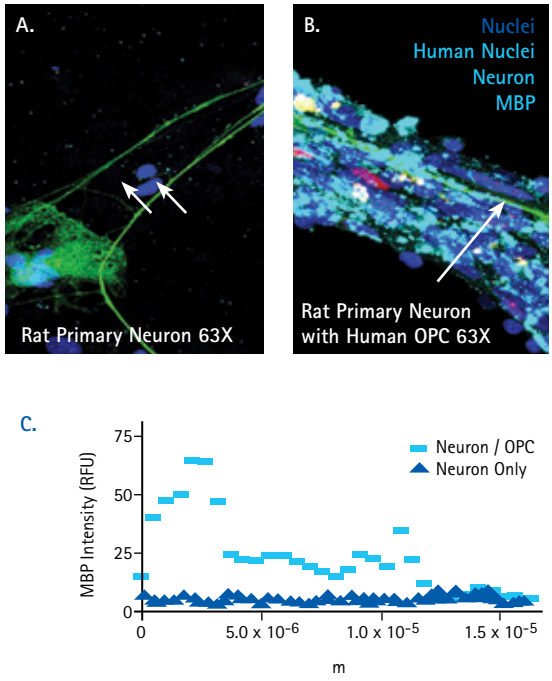
This panel of compounds consists of 80, well-characterized, cell permeable, potent and reversible protein kinase inhibitors targeting mainly CMGC and CaMK families of kinases; the majority of which are ATP-competitive. This library is useful for target identification in drug discovery, biochemical pathway analysis, screening new protein kinases, and other pharmaceutical-related applications. It is supplied with a CD containing comprehensive documentation for each inhibitor.

Graph (left): Screening of 160 kinase inhibitors included in InhibitorSelect™ libraries I and II. Data show Delta Confluence Values, corresponding to the change in relative cell number for twelve mock-treated wells and 160 kinase inhibitors. Three compounds, all affecting Rho kinases, were selected as primary hits for their effect on expansion of NES cells and are detailed in the top right.

Human Oligodendrocyte Differentiation Kit

(Catalogue No SCR600)

Easily generate enriched (>30%) populations of mature human oligodendrocytes in just 2-3 weeks with well-characterized oligodendrocyte progenitor cells (OPCs) and cell culture media for expansion and spontaneous differentiation. Included OPCs are guaranteed >70% GalC positive and Sox10 positive; mature oligodendrocytes are positive for MBP, PLP and MOG. Human OPCs can be used for studies of neurotoxicity, coculture applications and screening for inducers or inhibitors of preferential differentiation to mature oligodendrocytes.

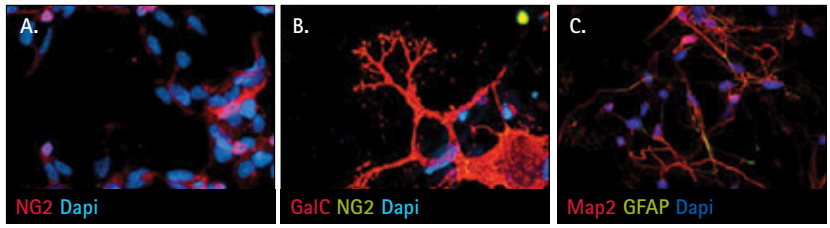


***In vitro* myelination assay.** Reconstructed 3D images of (Leica DMI 4000; 40x objective, 275x140 µm; 63x objective, 174.6 x 174.6 µm) rat primary neurons only (A) and rat primary neurons cocultured with human OPCs at 5:1 ratio for three weeks (B). All samples were stained with human-specific nuclei antibody (red) and myelin basic protein-specific antibody (cyan). MBP intensities (C) were measured by line scanning on cultures containing rat primary neurons only and rat primary neurons with human OPCs. GFP-labeled axons are indicated by the white arrow. The intensity of MBP greatly increased on the axons that were cocultured with human OPCs.

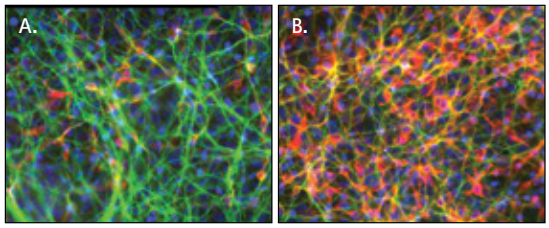
Human Oligodendrocyte Characterization Kit

(Catalogue No SCR601)

This is a convenient tool to fully evaluate human oligodendrocyte populations, and a great complement to the Oligodendrocyte Differentiation Kit. The kit contains markers to both early and late stage oligodendrocytes, including NG2, PLP, GalC, MOG, Map2, and GFAP.



Characterization of Merck Millipore's Human Oligodendrocyte Progenitor Cells (OPCs) (Catalogue No. SCR600) and their differentiated progenies. Proliferating Human OPCs express NG2 (A) After two weeks of spontaneous differentiation, approximately 30% cells become mature oligodendrocytes that express NG2, Gal C (B), while ~50% cells differentiate into MAP2 expressing neurons (C) with very little astrocytes (GFAP, C) detected. Human ES cell derived oligodendrocyte progenitor cells (Catalogue No. SCR600) were plated at 104/cm2 onto poly-L-ornithine (10 µg/mL) and laminin (10 µg/mL) coated 8 well chamber slides in Human OPC Expansion Complete Media (Catalogue No. SCM107). Twenty-four hours post-seeding, spontaneous differentiation was initiated by media exchange with Human OPC Spontaneous Differentiation Complete Media (Catalogue No. SCM106).

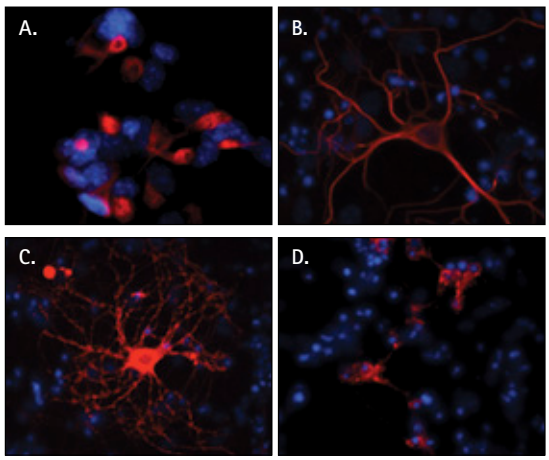


Human ES cell line, H9, derived neural progenitors were subjected to either spontaneous differentiation via growth factor withdrawal (A) or directed differentiation to dopaminergic neurons using SCR128 (B). Cells were fixed and stained with neuronal marker, MAP2 (MAB3418) and dopaminergic neuron marker, TH (AB152) at 1:400 dilution and counter stained with nuclei dye, DAPI. An elevated level of TH positive cells was observed when cells were differentiated using the Dopaminergic Differentiation Growth Factor Sampler Kit.

Dopaminergic Differentiation Growth Factor Sampler

(Catalogue No. SCR128)

Merck Millipore's Dopaminergic Differentiation Growth Factor Sampler contains a collection of five validated growth factors (Sonic Hedgehog (SHH), FGF-8, BDNF, GDNF, and TGFβIII) that are routinely used to induce differentiation of human pluripotent ES/iPS cells to dopaminergic neurons along with a reliable high affinity antibody marker, anti-tyrosine hydroxylase, to aid in quantifying the percentage yields from differentiation experiments.



Isolation, expansion and differentiation of fetal mouse primary hippocampal neuronal stem cells with serum-free medium containing supplement N21. Undifferentiated neural stem cells were stained with anti-nestin (A). Differentiated cells were stained with anti-MAP2 (B), CamKII (C) and O1 (D). All antibody staining is shown in red, and nuclei are visualized with DAPI staining.

N21 Medium Supplement

(Catalogue No. SCM081)

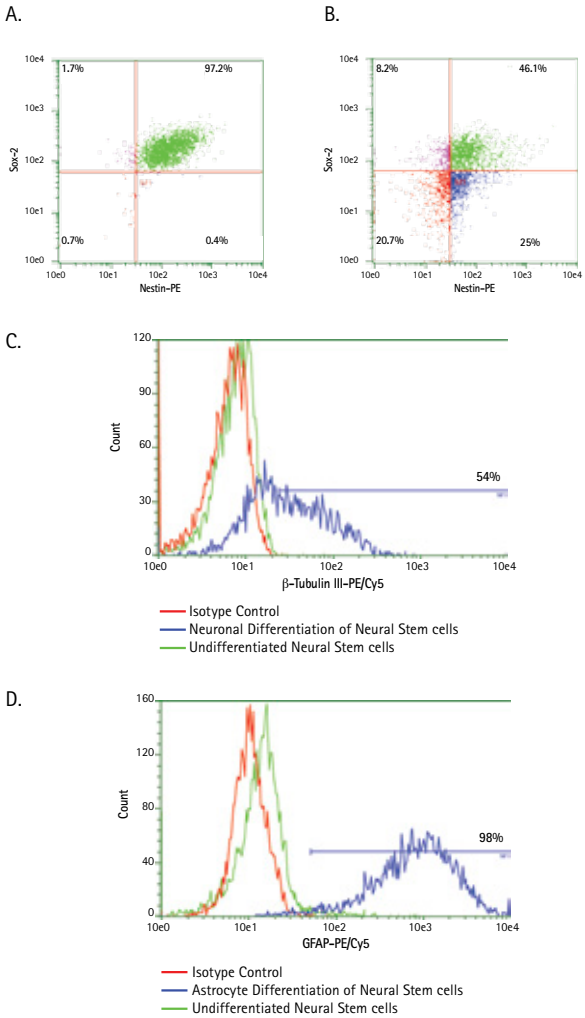
N21 medium supplement supports the survival and maturation of rat and mouse primary hippocampal neuronal progenitor cells under serum-free conditions. It may also be used to supplement formulations for the feeder-free, serum free culture of mouse embryonic stem (ES) cells and for the propagation and differentiation of human ES cell-derived neural progenitor cells. Stringent manufacturing processes and validation of N21 supplement minimizes the lot-to-lot variability found in other, similar serum-free supplements on the market.

FlowCellect® Rodent NSC Characterization Kit (Neuronal)

(Catalogue No. FCRNC25112)

The FlowCellect® rodent neural stem cell characterization kit is an easier and quicker way to track neuronal differentiation. The kit allows the researcher to calculate the percentage of undifferentiated stem cells in culture by determining the percentage of cells that express both Sox-2 and nestin as well as tracking βIII-tubulin up-regulation upon neuronal differentiation. This quick test allows researchers to test the quality of their cells in culture as well as measure changes in marker expression during differentiation.

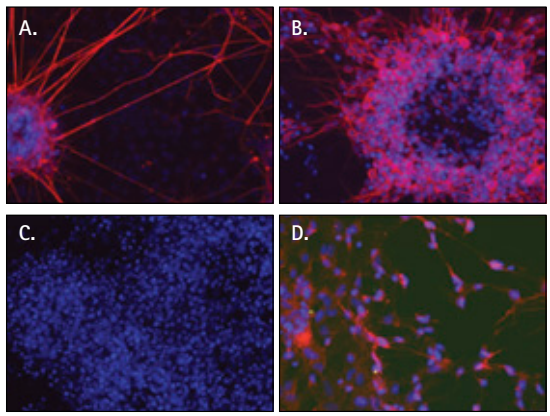
Graphs (right): Sox-2 and nestin are stem cell markers, while βIII-tubulin and GFAP are lineage markers. Before differentiation, NSCs are positive for nestin and Sox-2 (A), but these are down-regulated as differentiation begins (B). βIII-tubulin is up-regulated when cells are differentiated to neurons (C); likewise, GFAP is up-regulated when cells are differentiated to astrocytes (D).



ES2N Complete Medium Kit

(Catalogue No. SCM082)

ES2N complete medium is a defined, serum-free formulation that efficiently differentiates mouse ES and iPS cells into functional neurons. Traditional neuronal differentiation involves embryoid body (EB) formation in serum-containing medium. With the ES2N complete medium, cells readily differentiate into neuron monolayers within 9-12 days on gelatin-coated culture dishes, without the formation of EBs.



Immunocytochemistry analysis of mature neurons at day 12 differentiated from 129S6 mouse ES cells. (A). Mature neurons stained with tubulin antibody. (B). Mature neurons stained with MAP-2 antibody. (C). Antibody staining of the Oct4 pluripotency marker shows no Oct4 expression. (D). Antibody staining with the astrocyte marker GFAP. All stainings are overlayed with blue DAPI nuclear staining.

Key Products

For a complete listing of neuroscience products from Merck Millipore, please visit **www.millipore.com/neuroguide**.

Neural Stem Cell Antibodies

Description	Catalogue No.
Anti-BCRP, clone BXP-21	MAB4146
Anti-CD133, clone 13A4	MAB4310
Anti-Musashi-1	AB5977
Anti-Nanog, N-terminus	AB5731
Anti-Nanog	AB9220

Neural Stem Cells, Kits & Assays

Description	Catalogue No.
ENStem-A™ Human Neural Progenitor Expansion Kit	SCR055
ES2N Complete Medium Kit (includes ES2N Basal Medium plus Neuro27 and Neuro2 supplements)	SCM082
FlowCellect® Rodent NSC Characterization Kit (Neuronal)	FCRNC25112
Human Neural Stem Cell Characterization Kit	SCR060
Human Oligodendrocyte Differentiation Kit	SCR600
MilliTrace™ CX Nestin GFP Reporter Cell Line	SCR096
MilliTrace™ CX Constitutive GFP Reporter Cell Line	SCR095
MilliTrace™ VM Constitutive GFP Reporter Cell Line	SCR092
MilliTrace™ Constitutive GFP Reporter Adult Rat Hippocampal Neural Stem Cell Kit	SCR080
MilliTrace™ Nanog GFP Reporter Mouse Embryonic Stem Cell Kit	SCR089
Mouse Cortical Neural Stem Cells	SCR029
Mouse Spinal Cord Neural Stem Cells	SCR031
Neural Stem Cell Marker Characterization Kit	SCR019
N21 Medium Supplement (50X)	SCM081
Rat Hippocampal Neural Stem Cells	SCR022
ReNcell® VM Human Neural Stem Cell Kit	SCC010
ReNcell® CX Human Neural Stem Cell Kit	SCC009

Inhibitors

Description	Catalogue No.
InhibitorSelect™ 96-Well Protein Kinase Inhibitor Library III	539746
InhibitorSelect™ 384-Well Protein Kinase Inhibitor Library I	539743
StemSelect® Small Molecule Regulators 384-Well Library I	569744
Histone Acetyltransferase p300 Inhibitor, C646	382113
DNA Methyltransferase Inhibitor II, SGI-1027	260921

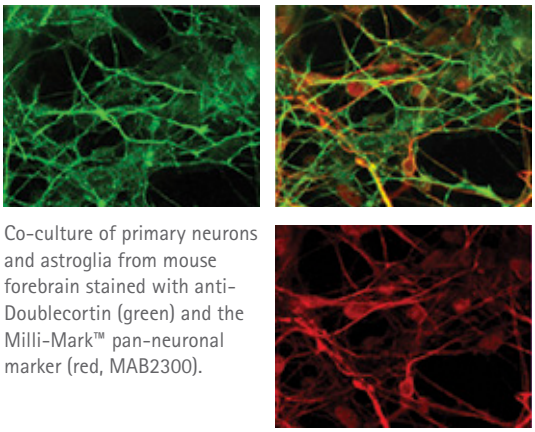
Neurite Outgrowth

Nervous system development results in a network of synaptic connections between participating neurons. Understanding the development of this network can improve therapeutics for nervous system developmental disorders and neurodegenerative disease. Neurons make connections by growing in response to axon guidance cues. Recent research has focused on how extracellular gradients affect asymmetric distribution and function of proteins driving neurite outgrowth. Merck Millipore's products for studying neurite outgrowth help determine the mechanisms of axon growth.

Anti-Doublecortin

(Catalogue No. AB2253)

Doublecortin (DCX) is a microtubule-associated protein used as a marker for neurogenesis because it is expressed almost exclusively in developing neurons. Neuronal precursors begin to express DCX shortly after exiting the cell cycle. They continue to express DCX for 2-3 weeks as the cells migrate and mature into neurons, at which point DCX is downregulated. In response to exercise, DCX expression increases in parallel with increased BrdU labeling, which is the traditionally accepted marker of neurogenesis.

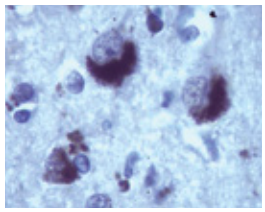


Co-culture of primary neurons and astroglia from mouse forebrain stained with anti-Doublecortin (green) and the Milli-Mark™ pan-neuronal marker (red, MAB2300).

Anti-Neurexin Antibodies

Neurexins are an intriguing group of heavily glycosylated and processed neuronal transmembrane surface proteins likely involved in cell recognition and adhesion and implicated in cognitive disease (Misslerr & Sudhoff 1998, TIG 14:20; Etherton et al. 2009, PNAS 106:17998). Merck Millipore introduced the first commercially available and highly validated antibodies to neurexins. Our Western blotting and immunohistochemistry data localize this protein to neuronal cytoplasm, consistent with previous research (Dean et al. 2003, Nat Neurosci 6:708-716).

Description	Catalogue No.
Guinea pig Anti-Neurexin-1-α	ABN35
Rabbit Anti-Neurexin-1-α	ABN98
Rabbit Anti-Pan-Neurexin 1	ABN161

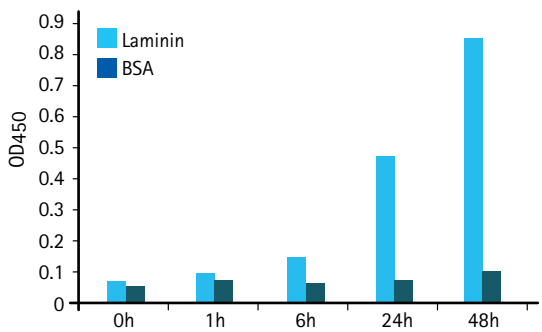


Paraffin-embedded human brain tissue Immunohistochemistry Analysis of ABN35, Anti-Neurexin-1-α.

Neurite Outgrowth Assay Plus Kit (3 µm)

(Catalogue No. NS230)

Merck Millipore's neurite outgrowth assay plus kit (3 µm pore size) uses Millicell® microporous tissue culture inserts, which contain permeable membranes that allow separation of neurite material from cell bodies for harvesting and use in this or other biochemical analyses. Induced neurites traverse these membrane pores to enable neurite quantitation or purification from the underside of the membrane.



Neurite extension on laminin-coated, but not BSA-coated, inserts dramatically increases over time.

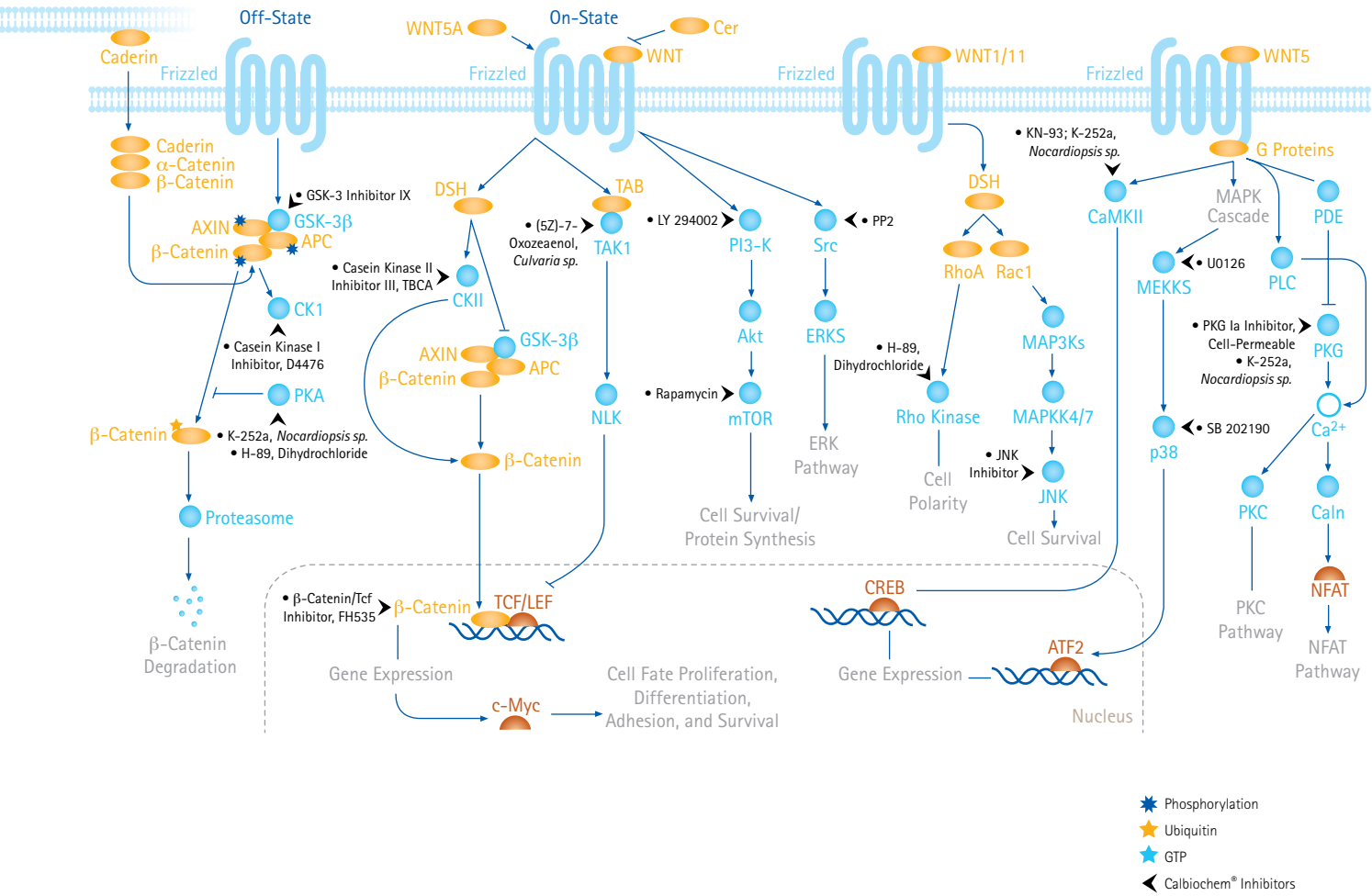
InhibitorSelect™ Wnt Signaling Pathway Inhibitor Panel

(Catalogue No. 681666)

The Wnt signaling pathway is an evolutionarily-conserved pathway involved in fate specification, development, cell proliferation, cell migration, and polarity, and migration of cells. Wnt genes encode a large family of secreted, cysteine-rich proteins that are also important in development and in maintenance of adult tissues.

Abnormalities in Wnt signaling are reported to promote both human degenerative diseases and cancer.

This panel contains 15 highly potent, selective, and cell permeable inhibitors (shown below in black) and a negative control for the investigation of the Wnt signaling pathway.



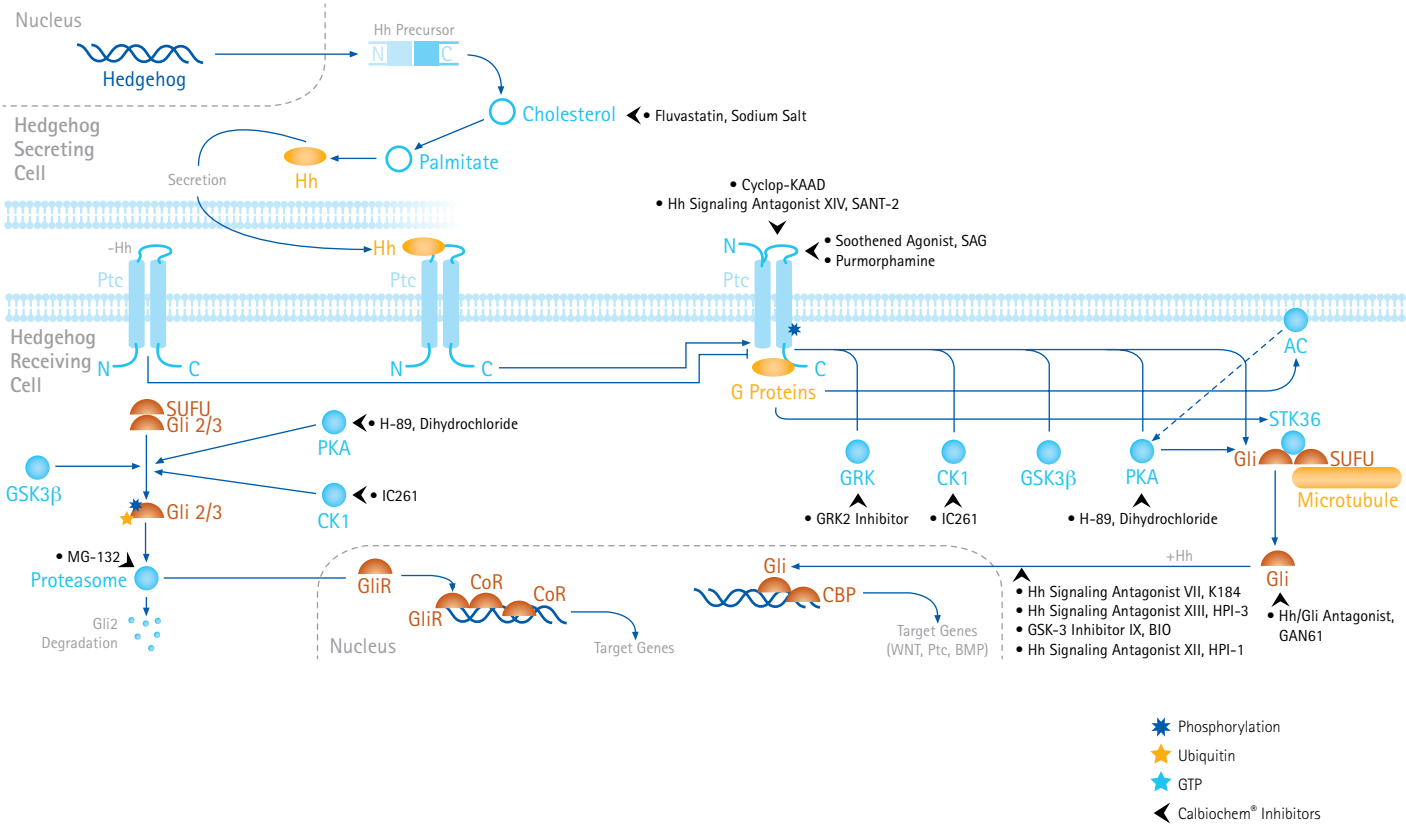
Hh Signaling Pathway Modulators

(Catalogue No. 373386)

Hedgehog signaling, a key intercellular signaling pathway in differentiation and organogenesis, is conserved from flies to human. Vertebrates express three different hedgehog proteins, of which Sonic hedgehog (Shh) is the best characterized. Shh has been implicated in several embryonic developmental processes. It displays inductive, proliferative, neurotrophic, and neuroprotective properties.

Mutations in the Shh pathway can lead to congenital defects and diseases, including cancer.

This panel consists of 14 potent, selective and cell-permeable antagonists, inhibitors and agonists (all shown below in black) and a negative control that are useful for the study of the Hh (Hedgehog) signaling pathway.



Key Products

For a complete listing of neuroscience products from Merck Millipore, please visit **www.millipore.com/neuroguide**.

Neurite Outgrowth Antibodies

Description	Catalogue No.
Anti-Doublecortin	AB2253
Anti-Neural Cell Adhesion Molecule	AB5032
Anti-Tubulin, βIII isoform, C-terminus, clone TU-20 (Similar to TUJ1)	MAB1637

Neurite Outgrowth Cell Culture Assays and ELISAs

Description	Catalogue No.
ChemiKine™ Brain Derived Neurotrophic Factor (BDNF) Sandwich ELISA Kit	CYT306
ChemiKine™ Nerve Growth Factor (NGF) Sandwich ELISA Kit	CYT304
Neurite Outgrowth Assay Kit (1 µm)	NS225
Neurite Outgrowth Assay Plus Kit (3 µm)	NS230
AXIS® Axon Isolation Device, tissue culture ready, 150 µm	AX15010TC
AXIS® Axon Isolation Device, tissue culture ready, 450 µm, Trial pack	AX45005TC
AXIS® Axon Isolation Device, tissue culture ready, 450 µm	AX45010TC
AXIS® Axon Isolation Device, tissue culture ready, 900 µm	AX90010TC
AXIS® Axon Isolation Device, tissue culture ready, 6-well	AX50010TC

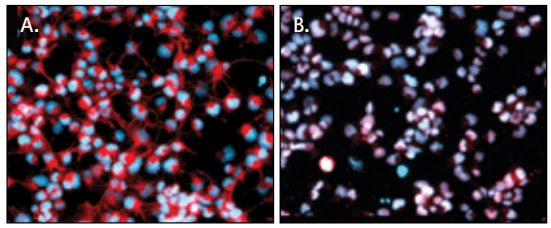
Extracellular Matrix (ECM)

ECM proteins regulate cell adhesion, differentiation, proliferation, migration, invasion, and survival. The expansive, delicate, malleable nature of neural architecture requires an intricate relationship with the ECM. Cultured neurons interact with laminin and fibronectin. Current studies ask whether these interactions are relevant for *in vivo* neurite degradation of and expansion through the ECM, synapse growth and withdrawal, dendritic arborization, and spine changes. Merck Millipore offers a wide variety of ECM antibodies, proteins, degradation assays and coated migration assays to meet each cell line's needs.

Synthetic Laminin Peptide

(Catalogue No. SCR127)

Laminin, an ECM protein, mediates cell adhesion, proliferation, migration, invasion, and differentiation. Merck Millipore's synthetic laminin peptide is a practical, convenient replacement for hard-to-purify native laminin, and is optimized to support adhesion, proliferation, and differentiation of rat NSCs.

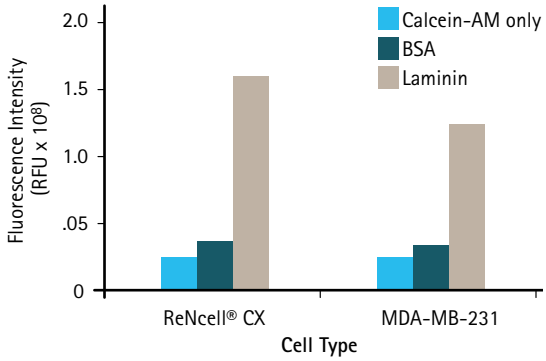


Rat NSCs cultured on synthetic laminin peptide-coated tissue cultureware express multipotent NSC markers, nestin (A, red) and Sox-2 (B, red). Cells were cultured for over 10 passages on synthetic laminin peptide-coated T-25 flasks (A,B).

QCM™ Laminin Migration Assay

(Catalogue No. ECM221)

Cell migration mediates embryonic development, wound healing, and immune response. Laminin promotes cell migration via integrin binding. Based on Boyden chambers, Merck Millipore's migration assays on laminin or fibronectin are accepted, effective tools to quantify the migration of neural, cancer, and stem cells.

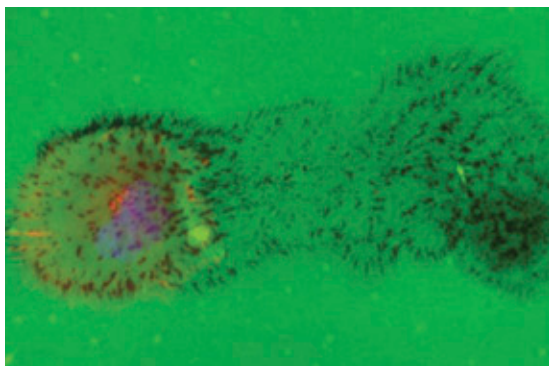


ReNcell® CX NSCs (SCC007, left) and breast cancer cells (right) demonstrate laminin-mediated migration (gray bars). Data are averages of triplicate samples.

QCM™ Gelatin Invadopodia Assays

(Catalogue Nos. ECM670 & ECM671)

Cell invasion into the surrounding extracellular matrix (ECM) is accompanied by the formation of actin-rich protrusions of localized protease activity, called invadopodia in cancerous cell types and podosomes in non-malignant cells. Merck Millipore's innovative QCM™ Gelatin Invadopodia Assays provide a simplified and standardized protocol for visualizing invadopodia in the context of nuclei and cytoskeleton. The assay kit includes all reagents for immunocytochemical analysis of cells migrating across a glass substrate coated with prelabeled fluorescein (green) or Cy3 (red) gelatin.



Cell migration across a fluorescently-conjugated matrix. Dark spots are evidence of invadopodia degradation. Cytoskeleton is stained with phalloidin (red) and nucleus is stained with DAPI (blue).

Key Products

For a complete listing of neuroscience products from Merck Millipore, please visit **www.millipore.com/neuroguide**.

ECM Proteins

Description	Catalogue No.
Synthetic Laminin Peptide	SCR127
Laminin, mouse purified	CC095
Human Plasma Fibronectin Purified Protein	FC010
Brain Derived Neurotrophic Factor, recombinant	GF029
NGF 2.5S, mouse	01-125
Ciliary Neurotrophic Factor, recombinant human	GF109
Platelet Derived Growth Factor-AB, recombinant human	GF106

ECM Assays

Description	Catalogue No.
ECMatrix™ Cell Invasion Assay, 96-well (8 µm)	ECM555
QCM™ Chemotaxis Assay 24-well (5 µm), Colorimetric	ECM506
QCM™ Chemotaxis Assay 96-well (5 µm), Fluorimetric	ECM512
QCM™ Haptotaxis Cell Migration Assay, Colorimetric	ECM580
QCM™ Gelatin Invadopodia Assay (fluorescent green)	ECM670
QCM™ Gelatin Invadopodia Assay (fluorescent red)	ECM671

Epigenetics and Gene Regulation

Why is epigenetics important? Neuronal signaling networks often converge on nuclear targets that induce transcription or repression of specific genes. In eukaryotic cells, genomic DNA is wrapped around histone protein complexes and compacted into dense chromatin fibres. Access to DNA, and the induction of transcription, replication, recombination and repair, is dictated by a complex code of epigenetic marks, including methylation of DNA bases, post-translational modifications to histone tails, and expression of variant histone proteins. These epigenetic marks mediate interactions between chromatin and remodeling enzymes and transcriptional complexes, and produce direct effects on gene regulation in neurons. Therefore, correlating epigenetic patterning with key neurological disorders and diseases may potentially reveal mechanisms of neural dysfunction.

DNA Methylation Analysis

5-methylcytosine (5-mC)

DNA methylation involves the addition of methyl groups to the 5′ carbon position of cytosine residues to form 5-methylcytosine (5-mC). This modification occurs mainly within the context of CpG dinucleotides, and is an important mechanism for chromosome stability, X chromosome inactivation, embryonic development, and gene silencing.

Detection of 5-mC by Bisulfite Modification

CpGenome™ Turbo Bisulfite Modification Kit

Bisulfite modification is an established technique for detecting methylated cytosines in genomic samples. In the bisfulfite reaction, all unmethylated cytosines are deaminated and sulfonated, converting them to uracils, while 5-methylcytosine remain unaltered. The CpGenome™ Turbo Bisulfite Modification Kit is designed to simplify and streamline the bisulfite modification process, enabling complete modification in as little as 90 minutes, starting from as little as 1 ng of DNA.

Description	Catalogue No.
CpGenome™ Turbo Bisulfite Modification Kit	S7847
CpGenome™ Fast DNA Modification Kit	S7824
CpGenome™ Universal DNA Modification Kit	S7820
Related Products	
CpG WIZ® APC Amplification Kit (human)	S7812
CpG WIZ® BRCA1 Amplification Kit (human)	S7830
CpG WIZ® Fragile X Amplification Kit (human)	S7807
CpG WIZ® GST-pi Amplification Kit (human)	S7808
CpG WIZ® hMLH1 Amplification Kit (human)	S7811
CpG WIZ® MGMT Amplification Kit (human)	S7803
CpG WIZ® Oct-4 Amplification Kit (mouse)	S7840
CpG WIZ® p14/ARF Amplification Kit (human)	S7817
CpG MethylQuest™ DNA Isolation Kit	17-10035
CpG MethylQuest™ Protein	14-921

Chromatin Immunoprecipitation (ChIP)

Gene expression is regulated by the interaction of various proteins with chromatin. Some of the most powerful investigations into epigenetic mechanisms of disease have utilized chromatin immunoprecipitation (ChIP). ChIP can detect and relatively quantify specific protein-DNA and protein-protein interactions *in vivo* at a single or multiple loci.

At the most elemental level, ChIP involves chemically crosslinking proteins to DNA sequences, which is followed by immunoprecipitation of the crosslinked complex, and final analysis of the resultant DNA by endpoint or quantitative PCR (qPCR), microarrays, or next-generation sequencing.

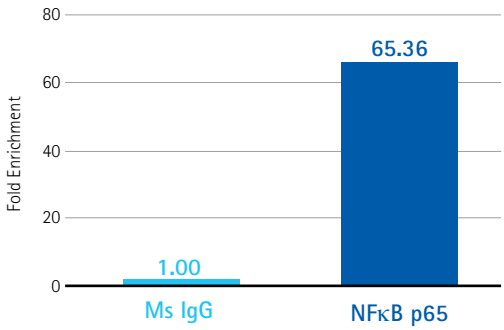
Since Upstate®, now part of Merck Millipore, launched the first ChIP kits in the 1990s, Merck Millipore has introduced an extensive line of ChIP technologies with many advantages:

- Improved sample prep
- One-day protocols
- High throughput ChIP
- Genome-wide analysis
- Tissue kits
- Optimized, specialized protocols
- Automation capability
- ChIP-validated antibodies
- Protein A, G and A/G beads

Magna ChIP® and EZ-Magna ChIP™ Kits

- Complete ChIP in just one day, from cells to PCR results
- Protein A/G magnetic beads; enrichment with wide range of antibodies
- Compatible with native ChIP (no cross-linking)
- EZ Magna kit with essential positive and negative control antibodies and qPCR primers

Specific localization of NFκB binding via one-day ChIP using the EZ-Magna ChIP™ kit. Sonicated chromatin prepared from serum-starved, TNFα-treated HEK293 cells (~3 x 10^6 cell equivalents per IP) were subjected to chromatin immunoprecipitation using 4 µg of either Normal Mouse IgG, or 4 µg Anti-NFκB p65 (RelA) (components contained in NFκB p65 ChIPAb+™ kit (Catalogue No. 17-10060).



Immunoprecipitation of NFκB p65 (RelA)-associated DNA fragments was verified by qPCR using primers directed against IκBα.

High Capacity Magnetic Beads for ChIP

Protein A, G, and A/G

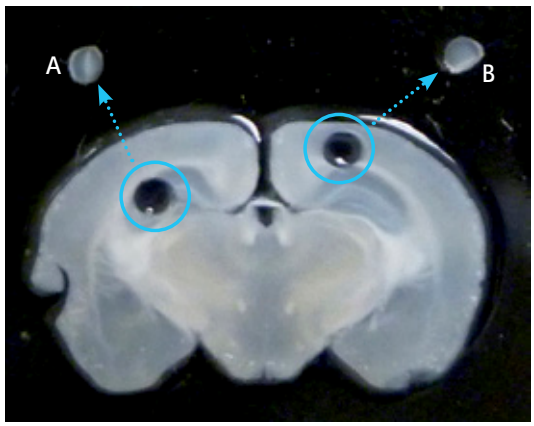
Magna ChIP® magnetic beads with protein A, G, or A/G are optimized specifically for ChIP applications and are a rapid, reproducible, and efficient reagent for collecting immunocomplexes in ChIP assays. Unlike conventional agarose beads, Magna ChIP® magnetic beads are rapidly moved to the side of a reaction vessel when exposed to a magnetic field, significantly reducing the handling time and mechanical stress on target immunocomplexes.

Description	Catalogue No.
Magna ChIP® A Kit	17-610
Magna ChIP® G Kit	17-611
EZ-Magna ChIP™ A Kit	17-408
EZ-Magna ChIP™ G Kit	17-409
Magna ChIP® A/G Kit	17-10085
EZ-Magna ChIP™ A/G Kit	17-10086
Magna ChIP® G Tissue Kit	17-20000
Magna ChIP® HT96 Kit	17-10077
EZ-Magna ChIP™ HT96 Kit	17-10078
Magna ChIP-Seq™ Chromatin Immunoprecipitation and Next Generation Sequencing Library Preparation Kit	17-1010
Magna ChIP2™ Universal Chromatin Immunoprecipitation DNA Microarray Kit	17-1000

Magna ChIP® G Tissue Kit

(Catalogue No. 17-20000)

Tissue-specific ChIP is a powerful method of elucidating mechanisms by which cell microenvironment can affect gene regulation. Conventional ChIP protocols work best using homogeneous populations of cultured cells. In contrast,this kit is specifically designed for tissue samples, which are more complex, heterogeneous mixtures of cells, and less amenable to ChIP. The Magna ChIP® G Tissue Kit provides the tools necessary to obtain repeatable, reliable, and site specific tissue biopsies. Microdissected, functionally-related populations of cells within a heterogeneous tissue can thus be analyzed with ease, precision and certainty.



Region-Specific Tissue Isolation. A 300 µm coronal mouse brain cryosection was obtained and two microdissections were carried out using the 1 mm microdissection punch provided in the kit. The isolated tissue is shown placed above the dissected region: (A) hippocampus, (B) cortex.

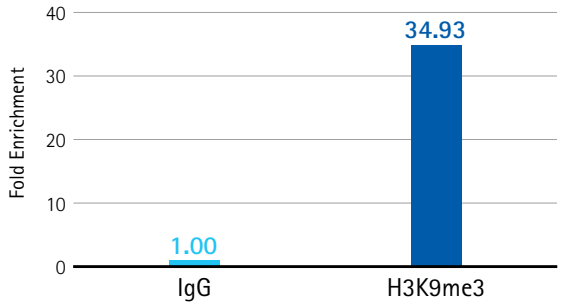
ChIP-validated Antibodies

ChIPAb+™ Kits

Avoid ChIP failure due to poor antibody performance by using ChIPAb+™ antibodies. To ensure reliable performance in your lab, each lot is individually validated and tested for ChIP. ChIPAb+™ kits are more than just an antibody. Each set also includes a negative control antibody, plus control primers for amplifying a known, enriched locus to help you validate your results.

Description	Catalogue No.
ChIPAb+™ Histone H2A.Z	17-10048
ChIPAb+™ Histone H2B	17-10054
ChIPAb+™ Histone H3 (C-term)	17-10046
ChIPAb+™ Histone H3 (Unmod Lys4)	17-675
ChIPAb+™ Acetyl Histone H3	17-615
ChIPAb+™ Acetyl-Histone H3 (Lys4)	17-10050
ChIPAb+™ Acetyl-Histone H3 (Lys9)	17-658
ChIPAb+™ Acetyl-Histone H3 (Lys14)	17-10051
ChIPAb+™ Monomethyl Histone H3 (Lys27)	17-643
ChIPAb+™ Dimethyl-Histone H3 (Lys4)	17-677
ChIPAb+™ Dimethyl-Histone H3 (Lys9)	17-648
ChIPAb+™ Trimethyl-Histone H3 (Lys4)	17-614
ChIPAb+™ Trimethyl-Histone H3 (Lys4)	17-678
ChIPAb+™ Trimethyl-Histone H3 (Lys9)	17-625
ChIPAb+™ Trimethyl-Histone H3 (Lys27)	17-622
ChIPAb+™ Trimethyl-Histone H3 (Lys36)	17-10032
ChIPAb+™ Trimethyl-Histone H3 (Lys79)	17-10130
ChIPAb+™ Phospho-Histone H3 (Ser10)	17-685
ChIPAb+™ Acetyl Histone H4	17-630
ChIPAb+™ Acetyl-Histone H4 (Lys5)	17-10045
ChIPAb+™ CREB	17-600
ChIPAb+™ CTCF	17-10044
ChIPAb+™ EED	17-663
ChIPAb+™ EED (Rabbit Polyclonal)	17-10034
ChIPAb+™ ERα	17-603
ChIPAb+™ EZH2, clone AC22	17-662
ChIPAb+™ HDAC1	17-608
ChIPAb+™ p53	17-613
ChIPAb+™ Phospho-CREB (Ser133)	17-10131
ChIPAb+™ REST	17-641
ChIPAb+™ RNA Polymerase II	17-620
ChIPAb+™ SMRT	17-10057
ChIPAb+™ Sox-2, clone 6F1.2	17-656
ChIPAb+™ Sp1	17-601
ChIPAb+™ SUZ12	17-661
ChIPAb+™ TATA Binding Protein (TBP)	17-10098

ChIPAb+™ trimethyl-histone H3 (Lys9)



ChIPAb+™+ trimethyl-histone H3 (Lys9) (17-625): Sonicated chromatin from NIH 3T3 L1 cells was subjected to chromatin immunoprecipitation using either normal rabbit IgG or Anti-trimethyl-histone H3 (Lys9) antibody and the Magna ChIP® A Kit (17-610). Successful enrichment of trimethyl-histone H3 (Lys9)-associated DNA fragments was verified by qPCR using primers flanking the mouse p16 promoter.

Magna ChIP-Seq™ Kit

- Reliable ChIP-Seq library construction from as little as 1 ng of purified ChIP DNA
- Protein A+G bead blend is compatible with a broader range of antibodies
- Flexible format allows construction of single end, paired end, or barcoded libraries
- Sufficient reagents for up to 10 next generation sequencing library constructions
- Quality-controlled, validated enzymes and buffers in convenient master mix streamline library construction
- Includes validated positive and negative control antibodies and a control primer set
- Proven performance through construction and sequencing of genomic DNA libraries on an Illumina Genome Analyzer II
- Expert support from our highly trained technical support scientists

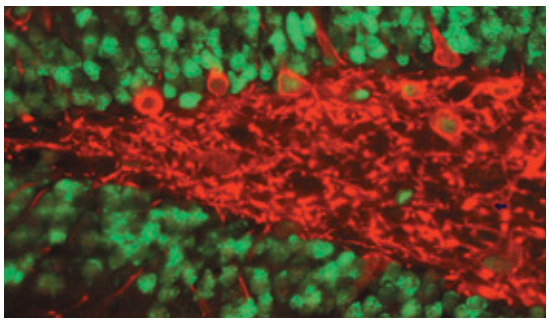
Description	Catalogue No.
Magna ChIP-Seq™ Chromatin Immunoprecipitation and Next Generation Sequencing Library Preparation Kit	17-1010

Differentiated Neurons

Neural stem cells can differentiate into three major cell types in the central nervous system: neurons, astrocytes, or oligodendrocytes. These building blocks of the brain must expand, connect, and mature to perform their sophisticated functions. The study of the differentiated nervous system includes many disparate fields, converging on key research areas described below, to which Merck Millipore is strongly dedicated.

Synapse and Structure

The differentiated nervous system is composed of an estimated 100 billion neurons and at least that many glial support cells. The diversity of neuronal and glial structure and function and the lack of distinguishing features often confound classification of newly differentiated cells in primary or slice cultures. The long, sinewy processes and delicate, branched structures of textbook neurons reflect their functions in processing and transmitting information. Merck Millipore is creating novel antibodies to neuron-specific proteins underlying this neural structure.



Mouse anti-NeuN (MAB377). Immunolocalization of NeuN (red) and BrdU (green) in the neurogenic regions of the mouse brain (dentate gyrus and subventricular zone). Photo courtesy of J.G. Emsley and T Hagg.

Neuronal Nuclei Marker: Anti-NeuN

Our exclusive NeuN antibody (clone A60) specifically recognizes the DNA-binding, neuron-specific protein NeuN, which is present in most CNS and PNS neuronal cell types of all vertebrates tested. NeuN is apparently restricted to neuronal nuclei, perikarya, and some proximal neuronal processes in both fetal and adult brain. NeuN antibodies are a standard in neuroscience laboratories and are cited in thousands of publications to date. Anti-NeuN is now available as a direct conjugate and as a rabbit polyclonal format.

Description	Catalogue No.
Anti-NeuN (guinea pig)	ABN90
Anti-NeuN (chicken)	ABN91
Rb Anti-NeuN polyclonal	ABN78
Rb Anti-NeuN – Biotin Conjugated	ABN78B
Rb Anti-NeuN – Alexa®488 Conjugated	ABN78A4
Rb Anti-NeuN - Cy3 Conjugated	ABN78C3
Ms Anti-NeuN monoclonal, clone A60	MAB377
Ms Anti-NeuN - Biotin Conjugated	MAB377B
Ms Anti-NeuN – Alexa®488 Conjugated	MAB377X

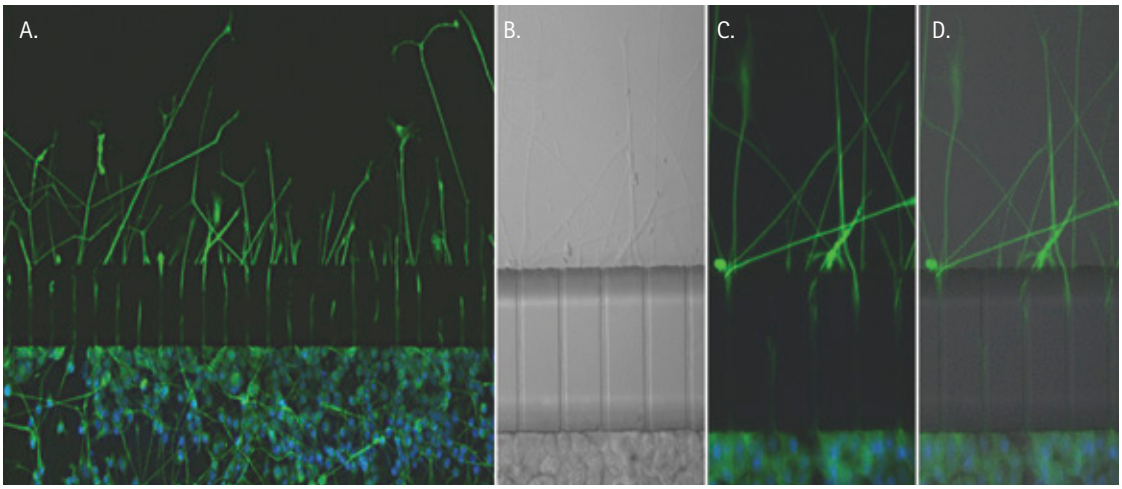
3 easy-to-use slide-based formats:

1. AXIS® Axon Investigation System

The AXIS® platform is Merck Millipore's most advanced tool for the study of neurite outgrowth. This slide-mounted microfluidic two-chamber system enables the deposition and culture of neural cells and the spatially controlled addition of growth factors, toxins, and other reagents. Neurite outgrowth is restricted to narrow, parallel channels, and the resultant outgrowth or collapse behavior is easily observed under a microscope. The result is a powerful platform for the study of somas, neurite outgrowth, and synapse formation.

Find more information at:
www.millipore.com/AXIS

Look for our application note, "**Spatially restricted exposure of toxins to neurons and its impact on axonal degeneration using AXIS®**" (Lit. No. AN4463EN00) at:
www.millipore.com/techlibrary



Images A-D show N1E-115 cells grown on an AX150 device. N1E-115 cells were loaded in the lower channel and cultured for 5 days in differentiation media. The cells were then fixed and stained with DAPI (blue) and with the neuronal cell stain MAB2300X (green).

A) fluorescent image showing the N1E-115 cells differentiating through the microgrooves of the AXIS® device. Note that the cell bodies (somas) are entirely contained on one side of the device and only the neurites are extended through the microgrooves into the other channel.

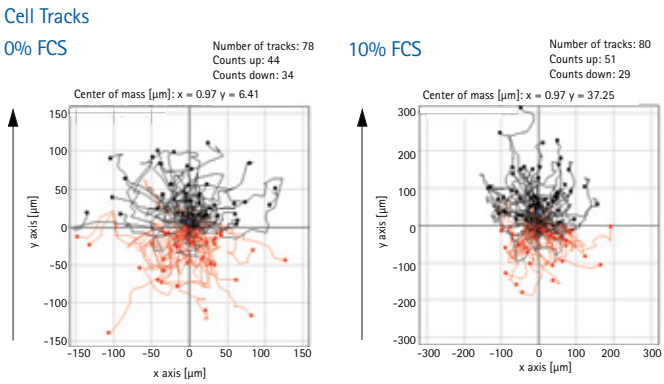
B) higher resolution white light image of the cells and device.

C) corresponding fluorescent image.

D) overlay of images B and C to verify that the neurites extend through the microgrooves only.

	AXIS® Axon Isolation Device Microgroove Length	Tissue Culture Ready (TC)	Plasma Bonded (PB)	Plasma Bonded to Confocal dishes (PBC)
Optimal Use		Ready-to-use devices. Just mount on slide and go. Good for slide or coverslip users.	Pre-sealed to glass slides to prevent leakage. Excellent for beginners.	Pre-sealed to coverslip glass culture dishes for confocal imaging from bottom.
Short experiments or maximal growth in axonal compartment	150 µm, 4 well	AX15010TC	AX15005PB	AX15005PBC
Most common axonal studies	450 µm, 4 well	AX45010TC	AX45005PB	AX45005PBC
More complex studies where 2 sets of microgrooves are needed	500 µm, 6-well	AX50010TC	AX50005PB	AX50005PBC
Longer axonal growth or transport studies	900 µm, 4 well	AX90010TC		

2. Millicell® µ-Migration Assay Kit



You won't get this kind of information with traditional migration assay platforms such as Boyden chambers or scratch (wound healing) assays. Data was analyzed and graphed using a free Image J software plug-in.

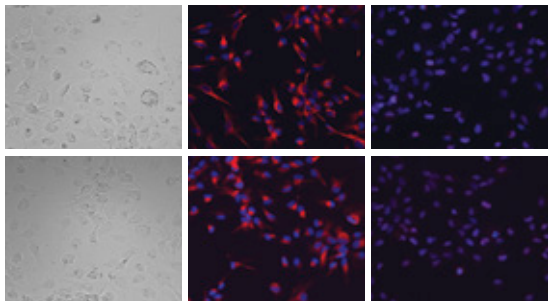
For detailed videos and instructions on the use of the µ-migration kit, please visit: **www.millipore.com/umigration**.

Measure the Effects of Chemoattractants on the Migration of Adherent Single Cells Through Real-Time Imaging

This unique platform makes it easy to measure the effects of chemoattractants or chemorepellants on the migration of adherent single cells with real-time imaging. A concentration gradient can be established for more than 48 hours; chemotaxis can be distinguished from random movement. The glass-like properties of the slide promote enhanced optical imaging for both slow- and fast-migrating cells thus allowing for multiparametric analysis for greater mechanistic insight. Parameters such as cell velocity, directionality and index can be quantified. Data is easily analyzed using free ImageJ plug-in software.

Description	Catalogue No.
Millicell® µ-Migration Assay Kit	MMA205

3. Millicell® EZ SLIDE



ReNcell® CX cells cultured on Millicell® 8-well glass EZ SLIDES (A, B, C) and Brand B 8-well glass chambered slides (D, E, F). Cells show staining for nestin (red, B and E) and Sox-2 (red, C and F). Nuclei (blue) are stained with DAPI.

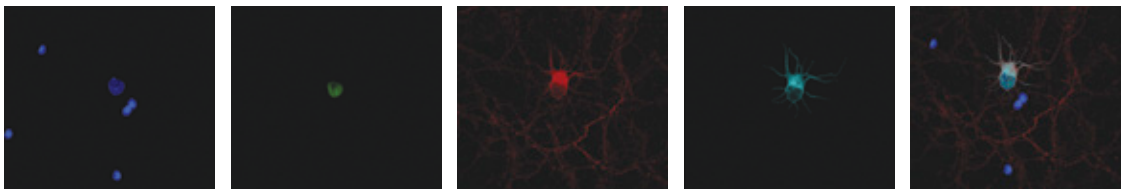
Simplify your cell analysis by using the Millicell® EZ SLIDE to culture, fix, stain and view your sample all in one device. There's no need to remove the medium chamber from the slide prior to fixing or staining. Unique, breakable tabs means you can easily remove wells without worrying about breaking slides or harming cells or messy glue or gasket residues. Acquire data simply and quickly with Millicell® EZ SLIDES.

Description	Catalogue No.
Millicell® EZ SLIDE (4-well glass, 16 pack)	PEZGS0416
Millicell® EZ SLIDE (4-well glass, 96 pack)	PEZGS0496
Millicell® EZ SLIDE (8-well glass, 16 pack)	PEZGS0816
Millicell® EZ SLIDE (8-well glass, 96 pack)	PEZGS0896
Millicell® EZ SLIDE Microscope Slide Holder	PEZXMSH01

New Neuro-Chrom™ Pan-Neuronal Markers

Superior Detection with Monoclonal Antibody Blends

Antibodies to neuronal proteins are critical tools for identifying neurons and discerning morphological characteristics. While Golgi staining and GFP constructs yield excellent cytoarchitectural detail, these pproaches are technically challenging and impractical for many neuroscience researchers. Neuron-specific antibodies reveal cytoarchitecture, but are limited by the protein distribution within the neuron. To achieve complete staining across all parts of neurons, Merck Millipore has developed a family of pan-neuronal antibody blends that label key somatic, nuclear, dendritic, spine, and axonal proteins. These pan-neuronal antibody cocktails have been validated in a variety of cell and tissue cultures, and are designed to offer researchers a convenient, specific, qualitative, and quantitative system for studying neuronal morphology.



DAPI | Nuclear Alexa Fluor® 488 dye | Somato-axonal Cy3 | Somato-dendric Alexa Fluor® 647 dye | Merge

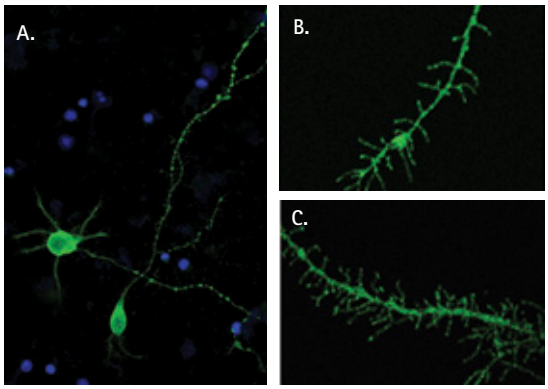
Catalogue No. NS420 was tested on rat cortex primary neurons (E18). Image was taken using a confocal microscope with appropriate filters. Fluorophore Alexa® 647 is represented in pseudo color. Cellular localization was scored with individual antibodies ranging from no (-) or minimal (+) to best (++++) staining.

Cellular localization	Alexa Fluor® 488 dye	Cy3	Alexa Fluor® 647 dye	Merge
Dendrites	-	++	++++	++++
Soma	-	++++	++++	++++
Nuclear	++++	-	-	++++
Axon	-	++++	+	++++

FluoroPan Neuronal Marker

(Catalogue No. MAB2300X)

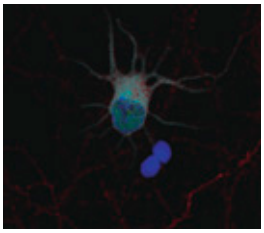
FluoroPan Neuronal Marker is an Alexa Fluor® 488 dye-conjugated monoclonal antibody blend, developed to enable detection of an entire neuron with only one microscopic emission channel. MAB2300X has been validated in diverse fixations, cell culture, and immunohistochemistry protocols, and is a convenient, specific, qualitative, and quantitative tool for studying neuronal morphology.



High morphological resolution has been achieved using MAB2300X including visualization of distal axons and dendrites with spines.

ChromaPan Neuronal Marker

(Catalogue No. NS420)



E18 rat cortical primary culture stained with NS420. Axons show in red; nucleus in green; and dendrites/soma in light blue. DAPI stain is also included in this kit.

The ChromaPan Neuronal Marker (NS420) is a complete multi-species neuronal antibody blend that specifically detects axons, dendrites, nuclei, and cell bodies with different fluorophores. As a result, different morphological features (somatic, nuclear, dendritic, and axonal) are illuminated in different colors.

Several additional kits have been developed to facilitate antibody colocalization studies within neuronal cultures. The NS330 and NS340 kits contain "open" channels for mouse and rabbit channels, respectively, providing the opportunity to choose additional antibodies to visualize neuronal expression of target proteins.

Description	Use	Catalogue No.
Pan Neuronal Marker – Unconjugated	Reseachers choose 2° antibody & can co-label using any non-mouse hosted antibodies.	MAB2300
FluoroPan Neuronal Marker – Alexa® 488 dye-conjugated	No 2° antibody needed! Researchers can co-label using any host.	MAB2300X
ChromaPan Neuronal Marker	Neuron parts stained in different colors. 1° antibody and 2° antibody blend makes simple protocol.	NS420
ChromaPan Neuronal Marker – Ms open (OMC)	No mouse host is used so researchers can co-label using their own mouse antibodies for the Alexa Fluor® 488 channel	NS330
ChromaPan Neuronal Marker – Rb open (ORC)	No rabbit host is used so researchers can use their own rabbit antibodies for the Cy3 channel.	NS340

Key Products

For a complete listing of neuroscience products from Merck Millipore, please visit **www.millipore.com/neuroguide**.

Synapse and Structure Antibodies & Small Molecules

Description	Catalogue No.
Anti-NeuN, clone A60	MAB377
Anti-Pan-Neuronal Neurofilament marker, clone SMI-311	NE1017-100UL
Anti-Neurofilament M (145 kDa), C-terminus	AB1987
Anti-Synaptophysin, clone SY38	MAB5258
Anti-PSD-95 AB9708	05-1308
Pan Neuronal Marker	MAB2300
FluoroPan-Neuronal Marker, Alexa Fluor® 488 conjugated	MAB2300X
ChromaPan Neuronal Marker	NS420
ChromaPan Neuronal Marker-OMC	NS330
ChromaPan Neuronal Marker-ORC	NS340
Neuronal Differentiation Inducer IV	480746
Neurogenesis Enhancer, P7C3A20	480744

Synapse and Structure Assays

Description	Catalogue No.
AXIS® Axon Isolation Device, Tissue Culture Ready, 150 µm	AX15010TC
AXIS® Axon Isolation Device, Tissue Culture Ready, 450 µm	AX45005TC
AXIS® Axon Isolation Device, Tissue Culture Ready, 450 µm	AX45010TC
AXIS® Axon Isolation Device, Tissue Culture Ready, 900 µm	AX90010TC
AXIS® Axon Isolation Device, Tissue Culture Ready, 6-well	AX50010TC

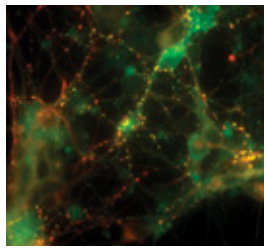
Neuroexcitability, Ion Channels, and Transporters

Synapses mediate neural excitation and modulate signal transmission. Not surprisingly, the vast majority of all neuropharmacological agents on the market today act at the synapse. Synaptic dynamics, receptor function, and neurotransmitter transport are heavily studied to gain insight into learning, memory, addictions, and many neurological diseases and disorders. Merck Millipore's antibodies from the expertise of Chemicon® have facilitated studies of the relationship between neurotransmitter transport, membrane depolarization, and neuroexcitability.

Anti-Vesicular Glutamate Transporter 1

(Catalogue No. AB5905)

VGLUT1 is expressed in a subset of glutamatergic neurons and transports glutamate into native synaptic vesicles. Anti-VGLUT1 mainly labels nerve fibers and terminals.

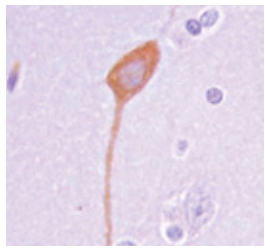


Localization of VGLUT1 (green) and synapsin (red) in rat brain hippocampal cells cultured for 7 days. Photo courtesy of QBMCell Science. QBMCellScience.com.

Anti-GAD67

(Catalogue No. MAB5406)

Glutamic acid decarboxylase is the enzyme responsible for the conversion of glutamic acid to GABA, the major inhibitory transmitter in higher brain regions, making GAD an excellent, well-published marker for inhibitory neurons.



Mouse anti-GAD67 (MAB5406) staining in mouse brain. Representative staining morphology in the CA1, CA2, CA3 (field of the hippocampus) and polymorph layer.

Key Products

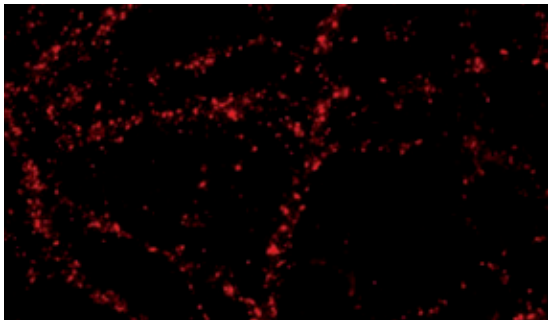
For a complete listing of neuroscience products from Merck Millipore, please visit **www.millipore.com/neuroguide**.

Neuroexcitability, Ion Channel, and Transporter Antibodies & Inhibitors

Description	Catalogue No.
Anti-Sodium Channel, Voltage Gated, Brain Type II	AB5206
Anti-Dopamine Transporter	AB15344
Anti-NMDAR1	AB9864
Anti-GluR1, N-terminus, clone RH95	MAB2263
Anti-GluR2, extracellular, clone 6C4	MAB397
Anti-Sodium Channel Nav1.8, pain	AB9274
Anti-Gad 65	ABN101
TrpA1 Antagonist, HC-030031	648485
NMDAR2C/2D Inhibitor, DQP-1105	454586
Kir2.1 Inhibitor, ML133	422689
Kir1.1 Inhibitor, VU591	422682
PICK1 PDZ Domain Inhibitor	529531

Cell Signaling

The diversity in neuronal receptors and channels reflects the complexity of signaling that drives neuronal development, excitability, and plasticity. From key phosphorylation and histone modification events in most neuronal processes, to calmodulin kinase regulation of synaptic plasticity and memory, to activation of AMPK pathways in hypothalamic energy regulation, signal transduction is a focus of research and a key to understanding normal and disease processes.

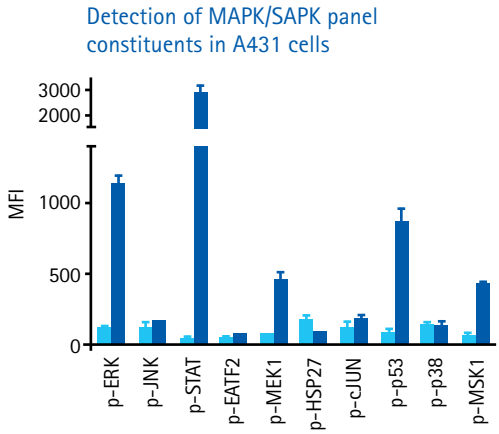


Rabbit anti-NMDAR2C (AB1592P) labels the surface of cultured hippocampal neurons.

Anti-NMDAR2C, also recognizes NMDAR2A and 2B

(Catalogue No. AB1592P)

N-methyl-D-aspartate (NMDA) receptors are a class of ionotropic glutamate receptors. The NMDA channel is involved in long-term potentiation, an activity-dependent increase in the efficiency of synaptic transmission thought to underlie certain kinds of memory and learning.



MILLIPLEX® MAP MAPK/SAPK Phosphoprotein Magnetic Bead 10 Plex Kit

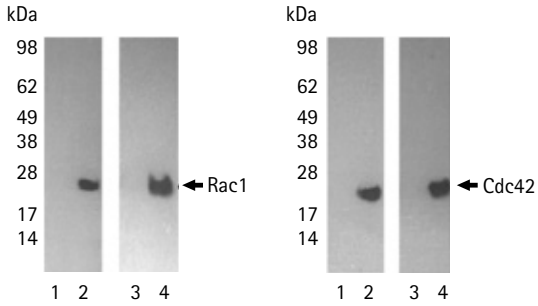
(Catalogue No. 48-660MAG)

Using the Luminex® xMAP® platform, our MILLIPLEX® MAP cell signaling portfolio enables quantitation of total or phosphorylated proteins involved in multi-pathway signaling or can be focused in a specific pathway. The MILLIPLEX® MAP MAPK/SAPK Phosphoprotein Magnetic Bead 10 Plex Kit can detect changes in phosphorylated Erk/MAP kinase 1/2 (Thr185/Tyr187), STAT1 (Tyr701), JNK (Thr183/Tyr185), MEK1 (Ser222), MSK1 (Ser212), ATF2 (Thr69/71), p53 (Ser15), HSP27 (Ser78), c-Jun (Ser73) and p38 (Thr180/Tyr182) in cell lysates.

Rac1/Cdc42 Activation Assay Kit

(Catalogue No. 17-441)

Rac1 and Cdc42 are small, GTP-binding signaling proteins that modulate cytoskeletal organization to affect cell migration, polarity, growth, and other processes. The Rac1/Cdc42 activation assay kit effectively detects Rac1 and Cdc42 activity in cell lysates by using the downstream effector of Rac1/Cdc42, p21-activated protein kinase (PAK1), to isolate the active, GTP-bound form of Rac1/Cdc42 from the sample.



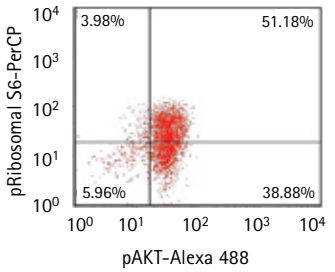
3T3/A31 (lanes 1, 2) and HeLa (lanes 3, 4) cell lysates were incubated with either GDP (lanes 1, 3) or GTPγS (non-cleavable GTP (lanes 2, 4). The lysates where then incubated with 10 µg of PAK1 PBD agarose to bind activated Rac1 (Rac1-GTP) and activated Cdc42 (Cdc42-GTP), which were detected by Western blotting with anti-Rac1 (left pair) or anti-Cdc42 (right pair). The arrows indicates Rac1-GTP and Cdc42-GTP, but not the GDP bound forms, bind to PAK1 PBD.

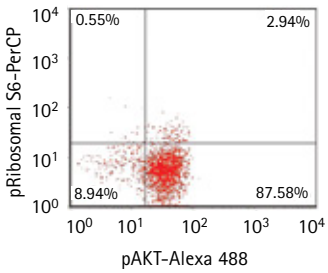
FlowCellect® PI3Kinase-mTOR Signaling Cascade Kit

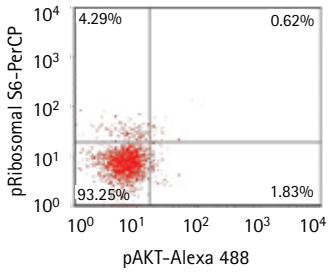
(Catalogue No. FCCS025210)

PI3 Kinase/mTOR signaling pathways regulate synaptic plasticity, memory retention, neuroendocrine regulation, and neuronal repair. FlowCellect® PI3K-mTOR analysis kit includes two directly conjugated phospho-specific mTOR pathway antibodies which are optimized for multi-color flow cytometry applications.

Graphs (left): PI3K pathway markers are activated (double positive population) in untreated Jurkat cells (A). After inhibitor treatment (wortmannin), all PI3K signaling biomarkers are inactivated (B). When using a specific inhibitor for mTOR activity alone (rapamycin), only phosphorylated ribosomal protein S6 is inactivated, with no effect on the levels of phospho-Akt (C).





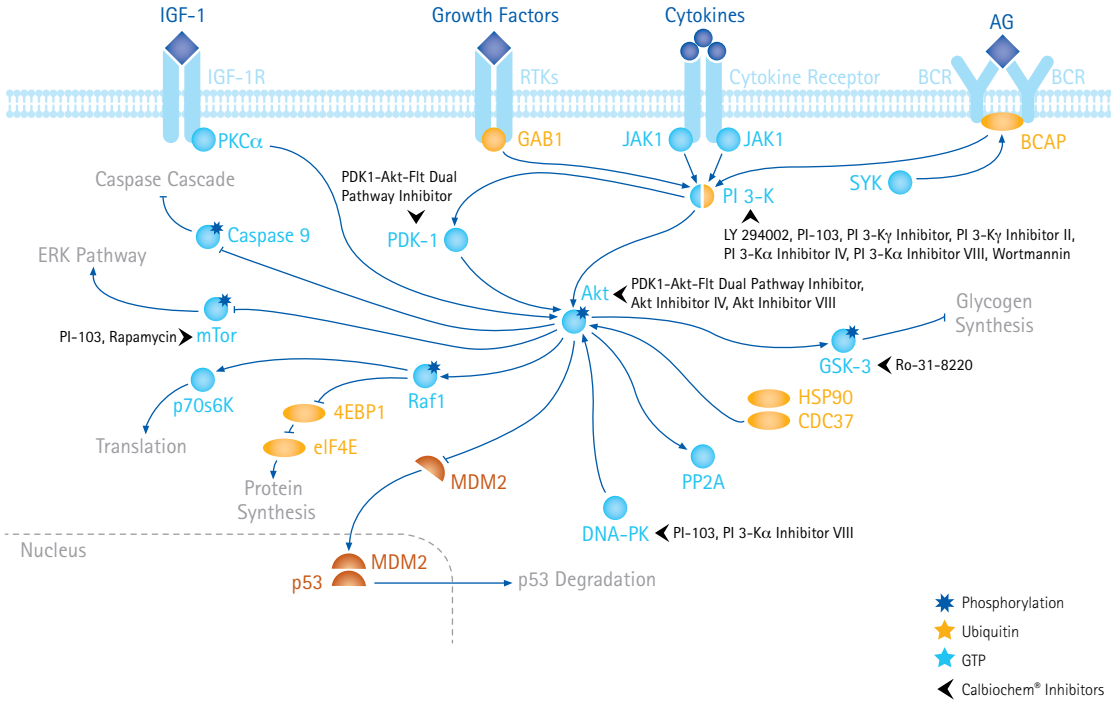


InhibitorSelect™ PI 3-K/Akt/mTOR Signaling Pathway Inhibitor Panel

(Catalogue No. 124031)

Activation of the PI 3-kinase/Akt/mTOR pathway stimulates cell proliferation and the translation process in response to nutrients and growth factors. This pathway regulates many processes of the central nervous system and is believed to play a key role in neurological disorders, such as Alzheimer's and Huntington's diseases.

This panel of 12 highly potent, selective, and cell-permeable Calbiochem® kinase inhibitors (shown below in black) and a negative control are useful for the investigation of the PI 3-K/Akt/mTOR signaling pathway. Each inhibitor is provided separately as a solid in glass vials. Each set also includes 15 mL of anhydrous DMSO. Each inhibitor in the panel, as well as the negative control, can also be purchased individually.



Key Products

For a complete listing of neuroscience products from Merck Millipore, please visit **www.millipore.com/neuroguide**.

Cell Signaling Antibodies

Description	Catalogue No.
Anti-NMDAR2C, also recognizes NMDAR2A and 2B	AB1592P
Anti-DARPP-32, phosphoThr34	AB9206

Cell Signaling Assays

Description	Catalogue No.
Cdc42 Activation Assay Kit	17-286
MILLIPLEX® MAP Human Akt/mTOR-11-plex Magnetic Bead Panel	48-611MAG
MILLIPLEX® MAP Human Total Akt/mTOR-11-plex Magnetic Bead Panel	48-612MAG
MILLIPLEX® MAP Multi-Pathway Magnetic Bead 9-Plex	48-680MAG
MILLIPLEX® MAP MAPK/SAPK Phosphoprotein Magnetic Bead 10 Plex Kit	48-660MAG
MILLIPLEX® MAP STAT Cell Signaling Magnetic Bead 5-plex Kit	48-610MAG
MILLIPLEX® MAP Phospho Akt1/PKB (Thr308) MAPmate™	46-645
MILLIPLEX® MAP Total CREB MAPmate™	46-632
MILLIPLEX® MAP Total Erk/MAP Kinase 1/2 MAPmate™	46-609
MILLIPLEX® MAP Total HSP27 MAPmate™	46-608
MILLIPLEX® MAP Total IRS1 MAPmate™	46-628
MILLIPLEX® MAP Total JNK/SAPK1 MAPmate™	46-618
MILLIPLEX® MAP Total STAT3 MAPmate™	46-625
MILLIPLEX® MAP Phospho STAT6 (Tyr641) MAPmate™	46-633
FlowCellect® PI3Kinase-mTOR Signaling Cascade Kit	FCCS025210
FlowCellect® PI3K/MAPK Dual Pathway Activation and Cancer Marker Detection kit	FCCS025100
FlowCellect® Multi-STAT Activation Profiling Kit	FCCS025550

Inhibitors

Description	Catalogue No.
Diisopropylfluorophosphate	30967
Tetrodotoxin, *Fugu sp.*	554412
CFTR Inhibitor-172	219670
Endothelin 1, Human and Porcine	05-23-3800
Adenylyl-imidodiphosphate, Tetralithium Salt	120002
nNOS –PSD-95 Interaction Inhibitor, ZL006	482740
Donepezil Hydrochloride	324377

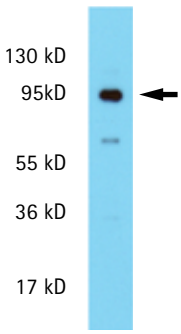
Sensory Systems and Metabolic Control

Sensing the environment and controlling body systems define an organism's behavior and are intricately tied to neural processes. For example, hypothalamic function maintains homeostasis, stress response, blood pressure, glucose levels, and satiety. Identification of key neurohormones and receptors has demonstrated a larger role for the CNS in adjusting parameters, such as energy expenditure, growth, and reproduction. Merck Millipore's comprehensive set of antibodies, assays, and multiplexing kits target soluble factors, receptors, and associated proteins involved in sensory systems and metabolism.

Anti-CLOCK

(Catalogue No. AB2203)

Circadian rhythm in most higher organisms is governed by key parts of the CNS, including the suprachiasmatic nuclei (SCN) of the hypothalamus, pineal gland, and retinal and extra-retinal receptor pathways. Circadian Locomotor Output Cycles Kaput, or CLOCK, is a transcription factor expressed in the SCN which regulates and is regulated by other circadian clock proteins. Polymorphisms within the CLOCK gene have been associated with circadian rhythm sleep disorders.

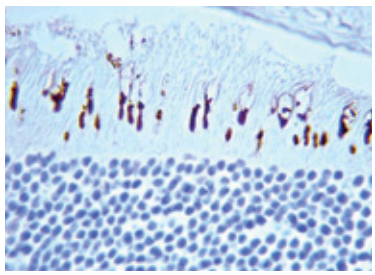


CLOCK protein is detected in L6 cell lysate by Western blotting with anti-CLOCK (AB2203).

Rabbit anti-Opsin, Red/Green

(Catalogue No. AB5405)

The full range of color discrimination in humans is based on the presence and function of three cone photoreceptor mechanisms. Each cone type possesses a photo-sensitive pigment-protein complex consisting of 11-cis retinal and a unique opsin protein, which gives sensitivity in the short (S cone, peak sensitivity about 420nm), middle (M cone, peak sensitivity about 530nm with polymorphism and long (L cone, peak sensitivity about 560nm with polymorphism wavelengths of the light spectrum.

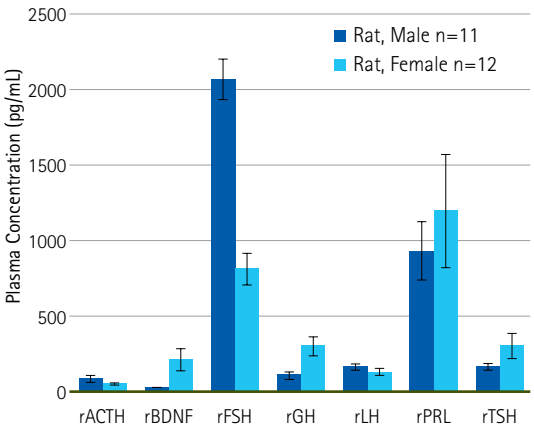


IHC staining of formalin-fixed, paraffin-embedded mouse retina using Rabbit anti-Opsin, Red/Green (Catalog No. AB5405).

MILLIPLEX® MAP Rat Pituitary Magnetic Bead Panel

(Catalogue No. RPTMAG-86K)

Called the "master gland" because it controls many other endocrine glands, the pituitary gland secretes a number of hormones that play important roles in the regulation of metabolism, growth, and reproduction. The Merck Millipore Rat Pituitary Magnetic Bead Panel enables you to explore various aspects of reproduction, growth, metabolic homeostasis, and pituitary-related diseases such as acromegaly, growth hormone deficiency, diabetes insipidus and pituitary tumors. Measure up to 7 key pituitary analytes (ACTH, BDNF, FSH, GH, LH, Prolactin, TSH) all from a single sample.



Quickly quantify differences in multiple biomarkers in each sample using the MILLIPLEX® MAP Rat Pituitary Magnetic Bead Panel. Male and female rats showed significant differences in plasma levels of BDNF, FSH and GH (error bars signify standard errors of the mean).

Also Available

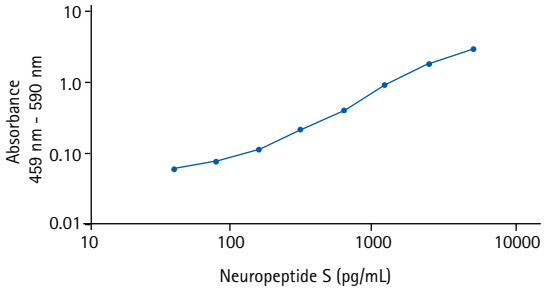
Description	Catalogue No.
MILLIPLEX® MAP Human Pituitary Magnetic Bead Panels 1 & 2	HPTP1MAG-66K
	HPTP2MAG-66K
MILLIPLEX® MAP Non-Human Primate Pituitary Magnetic Bead Panels 1 & 2	NHPPT1MG-46K
	NHPPT2MG-46K
MILLIPLEX® MAP Canine Pituitary Magnetic Bead Panel	CPTMAG-96K

Human Neuropeptide S (NPS) ELISA

(Catalogue No. EZHNPS-34K)

Neuropeptide S (NPS) is a neuropeptide that plays an important role in the regulation of metabolism and energy balance. It is produced mainly in the neurons in the amygdala, but neurons which are responsive to Neuropeptide S exist in many other areas of the brain. The Human Neuropeptide S ELISA kit is used for the quantification of Human Neuropeptide S in serum and plasma samples.

Typical Standard Curve for Neuropeptide S ELISA



Key Products

For a complete listing of neuroscience products from Merck Millipore, please visit **www.millipore.com/neuroguide**.

Sensory System and Metabolism Antibodies

Description	Catalogue No.
Anti-Arrestin, visual	MAB5580
Anti-Brain-Derived Neurotrophic Factor (BDNF)	AB1779SP
Anti-Capsaicin Receptor	MAB5568
Anti-Cocaine and Amphetamine Related Transcript	AB5340P
Anti-DARPP-32, phosphoThr75	AB9208
Anti-Dopamine Transporter, N-terminus, clone DAT-nt	MAB369
Anti-Ghrelin, Active	AB9756
Anti-Alpha7 Nicotinic Receptor, N-Terminus	AB5637
Anti-NMDAR2A and B	AB1548
Anti-Opsin, Blue	AB5407
Anti-Orexin-1 Receptor	AB3092
Anti-P2X3 Receptor	AB5896
Anti-mPER1 (residues 6-21)	AB2201
Anti-PER2	AB2202
Anti-PYY, amino acids 24-36	AB15666
Anti-Rhodopsin, clone C7	MAB2236
Anti-Substance P Receptor	AB5060

Sensory System and Metabolism Assays

Description	Catalogue No.
MILLIPLEX® MAP Human Pituitary Magnetic Bead Panel 1	HPTP1MAG-66K
MILLIPLEX® MAP Human Pituitary Magnetic Bead Panel 2	HPTP2MAG-66K
MILLIPLEX® MAP Human Neuropeptide Magnetic Bead Panel	HNPMAG-35K
MILLIPLEX® MAP Non-Human Primate Pituitary Magnetic Bead Panel 1	NHPPT1MG-46K
MILLIPLEX® MAP Non-Human Primate Pituitary Magnetic Bead Panel 2	NHPPT2MG-46K
MILLIPLEX® MAP Canine Pituitary Magnetic Bead Panel	CPTMAG-96K
ChemiKine™ Brain-Derived Neurotrophic Factor ELISA, Sandwich	CYT306
Chemikine™ Nerve Growth Factor ELISA	CYT304
Human Neuropeptide Y (NPY) ELISA	EZHNPY-25K
Rat/Mouse Neuropeptide Y (NPY) ELISA	EZRMNPY-27K
Human PYY (Total) ELISA	EZHPYYT66K
Human PYY (Total) RIA	PYYT-66HK
Human PYY (3-36) Specific RIA	PYY-67HK
Rat/Mouse PYY RIA	RMPYY-68HK

Degeneration

Diseases such as Parkinson's, Alzheimer's, amyotrophic lateral sclerosis (ALS), multiple sclerosis (MS), and prion diseases, as well as neurochemical degenerative factors like oxidative stress, free radical damage, and inflammation all follow complex cellular pathways. Merck Millipore offers a broad range of antibodies and detection kits for tracking many steps in neuronal degeneration.

Neurodegenerative Diseases/Disorders

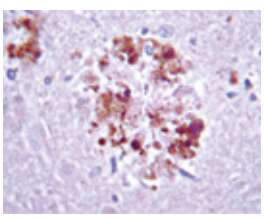
Neurodegenerative diseases are characterized by deterioration of neurons or their myelin sheath, disrupting transmission of sensory information, movement control and more. Because these cells are not easily regenerated, buildup of amyloid plaques can lead to disorders such as Alzheimer's and Parkinson's disease, and non-amyloid-related degeneration can cause ALS and MS. Merck Millipore offers validated biomarker antibodies and assays for elucidating the pathogenesis of both amyloid- and non-amyloid-related neurodegeneration.

Amyloid-Related Diseases

Anti-Alzheimer Precursor Protein A4, a.a. 66-81 of APP {N-terminus}, clone 22C11

(Catalogue No. MAB348)

Deposits of amyloid protein in senile plaques near nerve processes are found in the brains of aged humans and cases of Alzheimer's Disease. The principle component of this extracellular amyloid is β A4, a 4 kDa peptide derived from a larger amyloid precursor protein (APP), which is widely expressed in the brain and body.

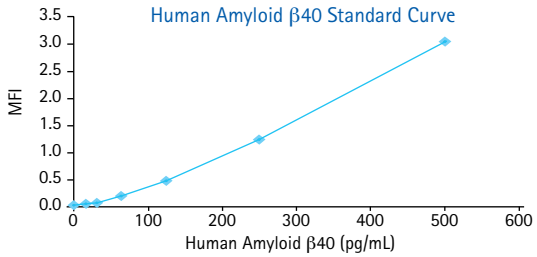


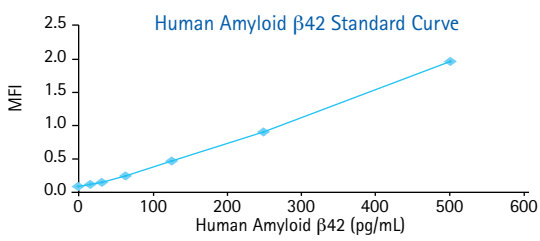
Anti-Amyloid β-A4-protein (MAB348) staining on Alzheimer's Disease-Hypothalamus

High Sensitivity Human Amyloid β40 & Amyloid β42 ELISA

(Catalogue Nos. EZHS40 & EZHS42)

Amyloid β peptides have been implicated in the etiology of Alzheimer's disease. Amyloid β40 is the most prominent peptide and Amyloid β42 is the neurotoxic form. Merck Millipore's High-Sensitivity (HS) ELISA Kits allow detection of as little as 6 pg/mL protein in cell culture lysates, primary neurons, lysates, plasma, and cerebral spinal fluid.





Quantifying Biomarkers in Neuroscience Research

Centuries of research have revealed the highly complex and interconnected nature of nervous system functions and exposed the need for the quantification of multiple neuro-specific biomarkers. ELISAs have long been employed for precise quantification of soluble targets from sera and lysates. Based on trusted ELISA principles, bead-based multiplex immunoassays have made possible multiparametric detection of analytes. Simultaneous measurement of key targets gives researchers a unique look at the concurrent processes that underlie neural development, homeostasis and pathogenesis. Merck Millipore is committed to developing innovative multiplexing kits for the study of neuroscience.

MILLIPLEX® MAP Human Neurodegenerative Disease Magnetic Bead Panels 1–3

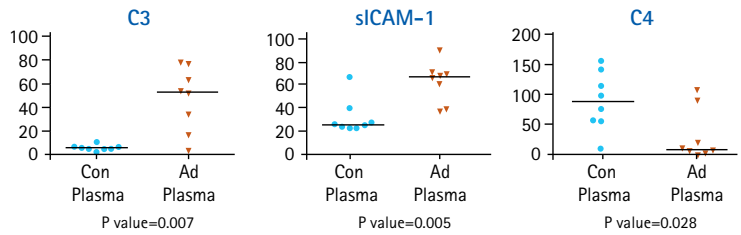
(Catalogue Nos. HNDG1MAG-36K, HNDG2MAG-36K, HNDG3MAG-36K)

Neurodegeneration is caused not only by targeted diseases such as Alzheimer's and Huntington's disease, but it can also be caused by inflammation or autoimmune disorders. Explore all the possibilities with Merck Millipore's neurodegenerative disease magnetic bead panels. These panels are designed to measure the levels of analytes in serum, plasma, CSF (cerebrospinal fluid), cell/tissue extract, or culture samples.

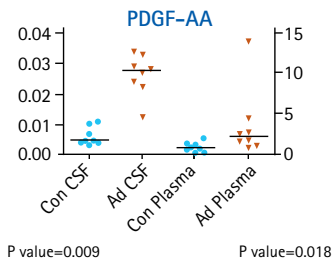
Analytes Available

Panel 1	α2-Macroglobulin, Apo AI, Apo CIII, Apo E, Complement C3, Complement Factor H, Prealbumin,
Panel 2	α1-Antitrypsin, Complement C4, CRP, MIP-4, PEDF, SAP,
Panel 3	BDNF, Cathepsin D, MPO, sNCAM, PAI-1 (total), PDGF-AA, PDGF-AB/BB, RANTES, sICAM-1, sVCAM-1

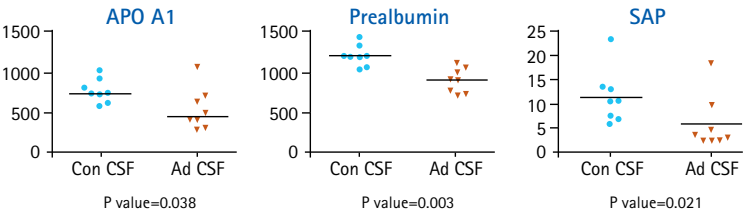
Relative Differences Between Normal and Alzheimer's Disease (AD) Samples: Plasma

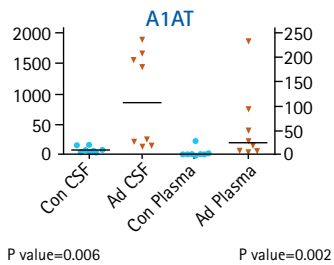


Relative Differences Between Normal and AD Samples: CSF and Plasma



Relative Differences Between Normal and AD Samples: CSF



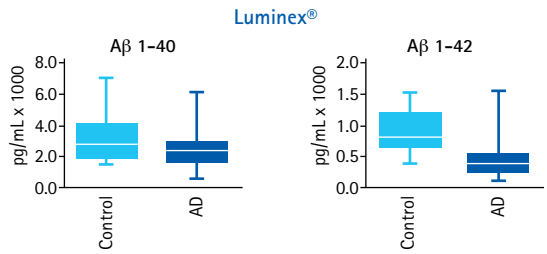


Merck Millipore's three human neurodegenerative disease magnetic bead multiplex kits (HNDG1-36K, HNDG2-36K and HNDG3-36K) were used to measure samples from both normal subjects and Alzheimer's disease samples. Significantly different biomarker levels were detected between these two groups.

MILLIPLEX® MAP Human Neurodegenerative Disease Magnetic Bead Panel 4

(Catalogue No. HNDG1MAG-36K)

Neurodegenerative diseases are progressive disorders that affect millions of patients. There is a need to identify biomarkers for diagnosis, prognosis and as surrogate markers for potential treatments. MILLIPLEX® MAP Human Neurodegenerative Disease Magnetic Bead Panel 4 is to be used for the simultaneous quantification of the following 5 analytes in any combination in diluted human cerebrospinal fluid: Amyloid β1-40, Amyloid β1-42, GDNF, sRAGE and S100B.



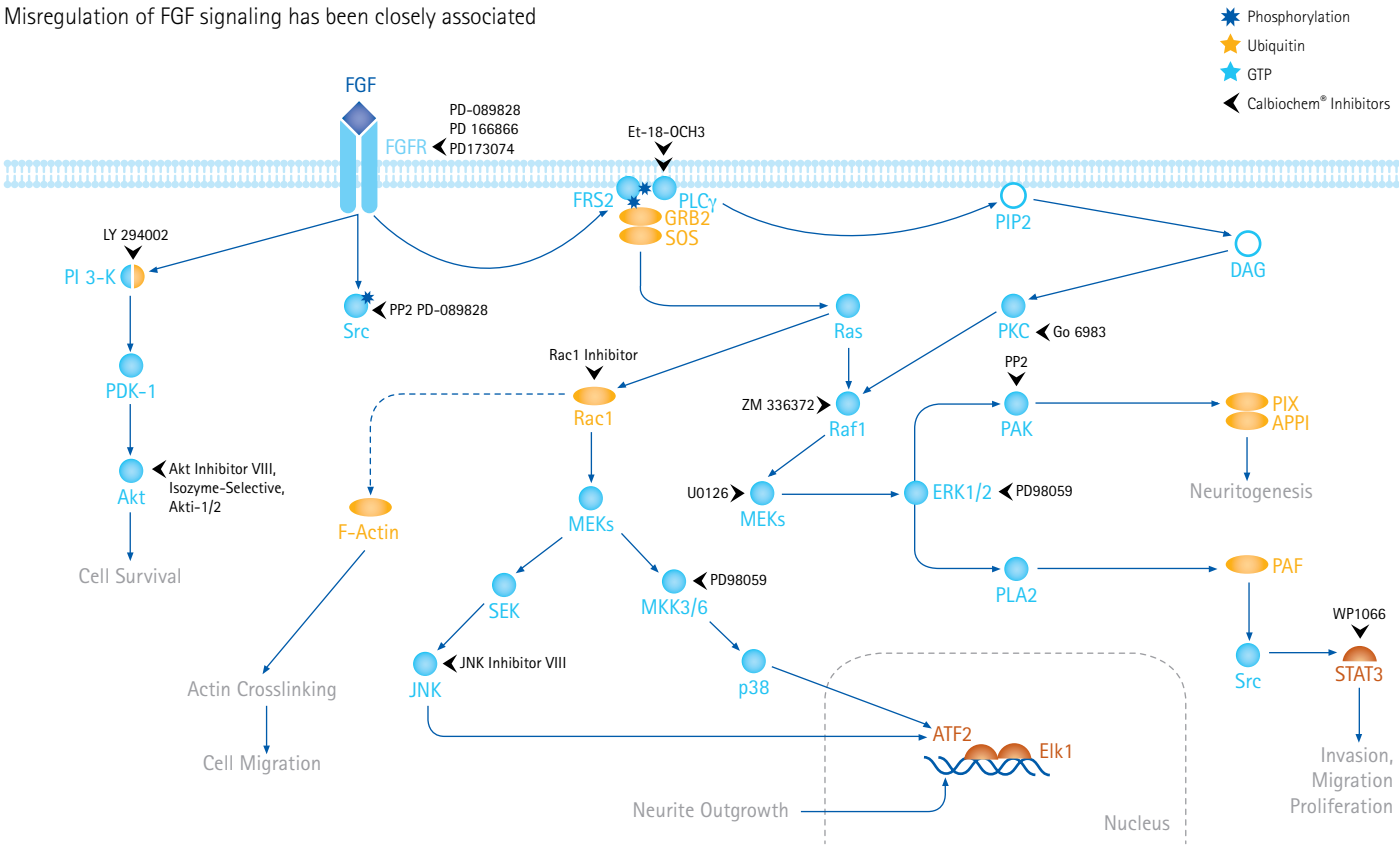
Levels of Aβ 1-40 and 1-42 in CSF obtained from AD patients and age-matched controls using MILLIPLEX® MAP Neurodegenerative Disease Magnetic Bead Panel 4. Aβ (1-40 and 1-42) values were determined using 16 AD and 16 age-matched control CSF samples. The median values for MILLIPLEX® MAP assays were control=2824 pg/mL and AD=803 pg/mL and control=2351 pg/mL and AD=387 pg/mL for Aβ(1-40) and Aβ(1-42), respectively.

InhibitorSelect™ FGF Signaling Pathway Inhibitor Panel

(Catalogue No. 341612)

Fibroblast Growth Factors (FGF) are heparin-binding proteins, which interact with cell-surface associated heparin sulfate proteoglycans to produce a wide array of cellular and physiological effects, such as angiogenesis, wound healing, and embryonic and neural development. Misregulation of FGF signaling has been closely associated with various human diseases, such as Michel aplasia, neoplasia, Parkinson disease, Pfeiffer syndrome and hereditary spinocerebellar ataxias.

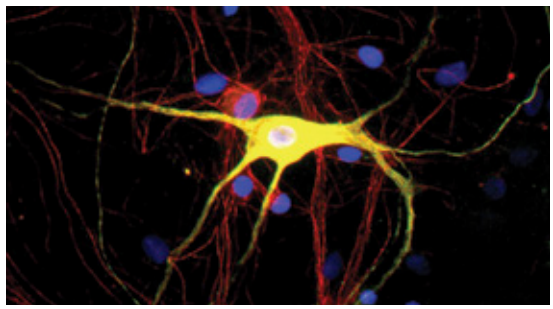
This panel consists of 14 potent, selective, reversible and cell-permeable inhibitors (shown below in black) and a negative control for the study of multiple signaling pathways activated by FGF.



Anti-Peripherin

(Catalogue No. AB9282)

Mutations in the peripherin gene have been correlated with incidence of ALS; therefore, studying its localization and function can help to elucidate ALS pathophysiology at a molecular level. Merck Millipore's anti-perpherin antibody is validated for immunocytochemistry and Western blotting.

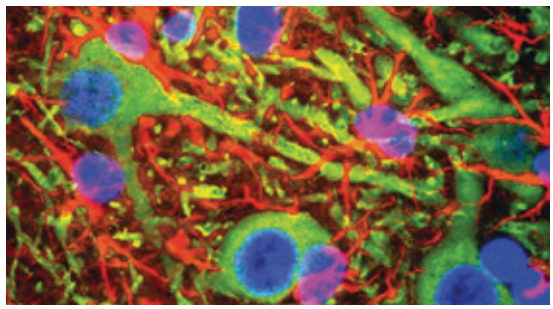


Localization of peripherin (Catalogue No. AB9282, yellow) and alpha internexin (Catalogue No. AB5354, red) in a p18 rat derived brain cortical neuron/glial mixed culture.

Anti-GFAP, clone GA5

(Catalogue No. MAB360)

Glial fibrillary acidic protein (GFAP) makes up the intermediate filaments in astrocytes. It is a cell-specific marker that distinguishes differentiated astrocytes from other glial cells, making it useful for studying degeneration of the central nervous system. This monoclonal antibody is validated for ICC, IHC, and Western blotting.

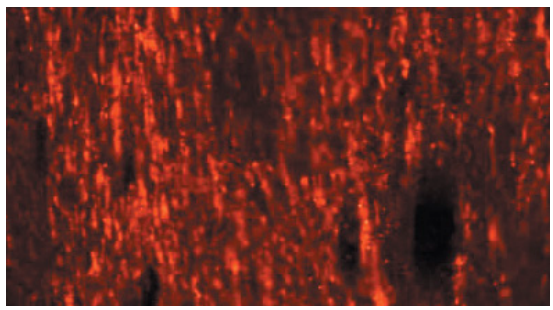


Immunolocalization of MAP2 (Catalogue No. AB5622, green) and GFAP (Catalogue No. MAB360, red) in rat hippocampus. Photo courtesy of Karl A. Kasischke & Patricia J. Fisher, Cornell University, Ithaca, NY.

Anti-Myelin Basic Protein (MBP)

(Catalogue No. AB5864)

The protein encoded by the classic MBP gene is a major constituent of the myelin sheath of oligodendrocytes and Schwann cells in the nervous system. MBP proteins are responsible for forming and stabilizing the myelin membranes of the CNS. However, alternative splicing using upstream exons creates MBP-related transcripts that function in the bone marrow and the immune system, inducing T-cell proliferation.



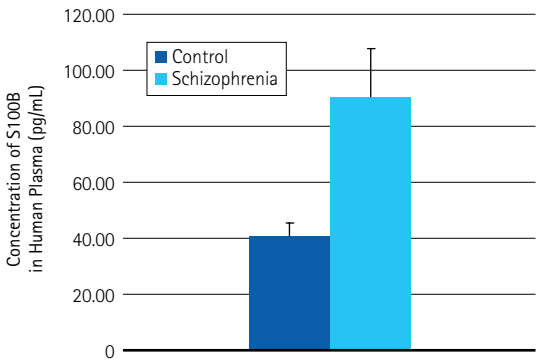
Rabbit anti-degraded myelin basic protein (Catalogue No. AB5864) staining of distal segment of ligated sciatic nerve.

Human S100B ELISA

(Catalogue No. EZHS100B-33K)

S100B is produced by astrocytes in the central nervous system and has been implicated in the development and maintenance of the nervous system. Although the mechanism of S100B secretion is unknown, it is affected by oxidative stress. At normal levels, S100B protects neurons against glutamate toxicity. Glial damage or astrocytic reactions to neural injury (reactive astrogliosis) causes an increase in S100B. High levels of S100B may indicate damage or dysfunction of CNS. This kit is designed for quantification of human S100B in CSF, serum and plasma.

S100B Expression Levels in Study Groups



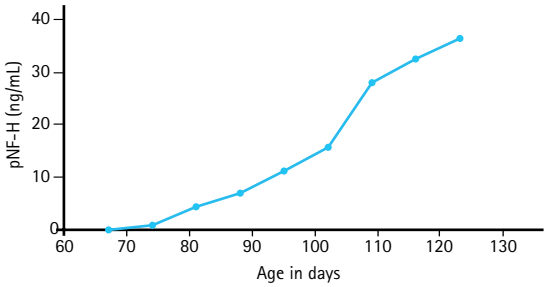
Plasma from Schizophrenia patients shows higher expression levels of S100B compared to control subjects. S100B was measured in plasma using the Human S100B ELISA (Catalogue No. EZHS100B-33K). The sample set was split into two groups (control n=81, Schizophrenia n=80). A non-parametric test (Mann-Whitney) was applied to compare mean values between both groups. A significant difference is observed in the mean values between control and the Schizophrenia group (p=0.003, error bars indicate the standard error of the mean).

Phosphorylated Neurofilament, (pNF-H) Sandwich ELISA

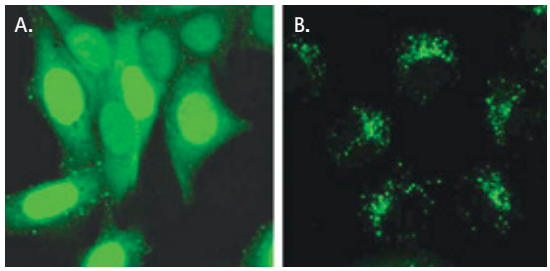
(Catalogue No. NS170)

Neurofilaments make up the major part of the cytoskeleton of neurons and are particularly concentrated in larger diameter axons. The phosphorylated forms of the heavy subunit (NF-H) are also quite resistant to proteases, suggesting that this very abundant protein might be particularly resistant to proteotytic cleavage upon release from damaged and diseased axons. Merck Millipore's phosphorylated neurofilament sandwich ELISA is sensitive to approximately 0.0585 ng/mL.

Blood pNF-H in G93A SOD1



Measurement of pNF-H using NS170 in transgenic mouse for human copper/zinc superoxide dismutase 1 (SOD1) (incorporated G93A mutation). This mutant SOD1 (found in some familial forms of ALS) causes a disease state in the mouse very similar to human ALS. The blood was taken by tail bleeding, and 0.5 microliters of plasma was used for the assay. There is a weak signal in most mice at 74 days which increases as the disease progresses. The animals do not show obvious ALS symptoms until about 90 days, so the assay can clearly pick up presymptomatic axonal loss.



HeLa cells were plated in a chamber slide and transduced with lentiviral particles. Cells were either left in complete media (A) or incubated in EBSS with a lysosomal inhibitor (B) to induce autophagy and inhibit lysosomal degradation of autophagosomes. GFP-LC3 is seen to aggregate in autophagosomes after treatment.

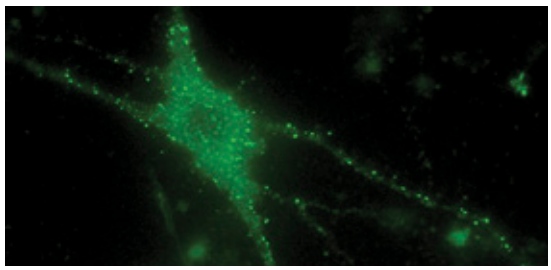
Watch the video of GFP-LC3 localizing to the autophagosome in live cells.

LentiBrite™ GFP-LC3 & GFP-LC3 Mutant Lentiviral Biosensors

(Catalogue Nos. 17-10193 & 17-10189)

Autophagy, a degradative pathway that provides recycled nutrients to cells under stress, plays both protective and deleterious roles in many diseases, including cancer, neurodegeneration, and infections. Upon induction of autophagy, LC3 (the mammalian homolog of the small protein Atg8) is conjugated to phosphatidylethanolamine and recruited to the autophagosome membrane, targeting the autophagosome for fusion to the lysosome. Merck Millipore's LentiBrite™ GFP-LC3 and mutant GFP-LC3 Lentiviral Biosensors provide bright fluorescence and precise localization to enable live cell analysis of autophagy in difficult-to-transfect cell types.

Visit **www.millipore.com** for a complete list of LentiBrite™ lentiviral biosensors.

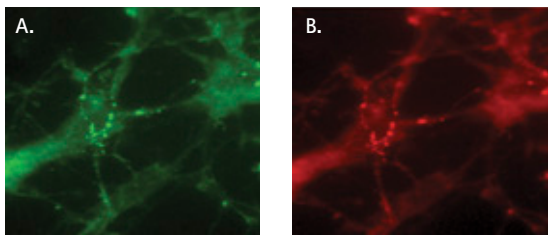


Primary rat hippocampal neuron cells were plated in a chambered cover glass and transduced with LentiBrite™ PSD95-GFP lentiviral particles. After media replacement and 2 weeks further incubation, cells were imaged live by oil immersion wide-field fluorescence microscopy. The PSD95-GFP displays a dotted distribution along the neurites.

LentiBrite™ PSD95-RFP & PSD95-GFP Lentiviral Biosensors

(Catalogue Nos. 17-10226 & 17-10227)

MAGUKs (membrane-associated guanylate kinases) serve both as central organizers of the postsynaptic density (PSD) and as signaling molecules. One such MAGUK, PSD-95, is associated with the dendritic spines of mature synapses, and is relatively immobile compared to other MAGUKs. PSD-95 fused to fluorescent proteins has been employed to quantify turnover and mobility of PSD-95. Merck Millipore's LentiBrite™ PSD-95-GFP & -RFP lentiviral particles provide bright fluorescence and precise localization to enable live cell analysis of PSD dynamics in neuronal synapses.



Immunocytochemistry comparison of rat hippocampal neuron cells transduced with LentiBrite™ PSD95-GFP lentiviral particles. ICC staining (red) (B) of the same fields of view with a monoclonal antibody against PSD95 (Cat No. MAB1596) reveals similar expression patterns to the GFP-protein (green) (A).

Key Products

For a complete listing of neuroscience products from Merck Millipore, please visit **www.millipore.com/neuroguide**.

Amyloid Biomarker Antibodies, Proteins, and Assays

Description	Catalogue No.
Alzheimer's Disease	
Anti-β-amyloid 1-42	AB5078P
Anti-pan amyloid β peptide (MOAB2), clone 6C3	MABN254
Anti-Alzheimer Precursor Protein A4, a.a. 66-81 of APP (N-terminus), clone 22C11	MAB348
Anti-Amyloid Fibril OC	AB2286
Anti-Amyloid Fibril LOC	AB2287
Anti-Tau phospho Threonine 231	AB9668
Anti-Human Amyloid β, clone WO-2	MABN10
Anti-Human Amyloid β40, clone G2-10	MABN11
Anti-Human Amyloid β42, clone G2-11	MABN12
Anti-Human Amyloid β42, clone G2-13	MABN13
Human S100B ELISA	EZHS100B-33K
High Sensitivity Human Amyloid β40 ELISA	EZHS40
High Sensitivity Human Amyloid β42 ELISA	EZHS42
High Sensitivity Human Amyloid β40 and Amyloid β42 ELISA	EZHS-SET
Human Amyloid β40 Brain ELISA	EZBRAIN40
Human Amyloid β42 Brain ELISA	EZBRAIN42
Human Amyloid β40 and Amyloid β42 Brain ELISA	EZBRAIN-SET
MILLIPLEX® MAP Human Neurodegenerative Disease Magnetic Bead Panel 1	HNDG1MAG-36K
MILLIPLEX® MAP Human Neurodegenerative Disease Magnetic Bead Panel 2	HNDG2MAG-36K
MILLIPLEX® MAP Human Neurodegenerative Disease Magnetic Bead Panel 3	HNDG3MAG-36K
MILLIPLEX® MAP Human Neurodegenerative Disease Magnetic Bead Panel 4	HNDG4MAG-36K
MILLIPLEX® MAP Human Neurological Disorders Magnetic Bead Panel 1	HND1MAG-39K
MILLIPLEX® MAP Human Neurological Disorders Magnetic Bead Panel 2	HND2MAG-39K
Huntington's Disease	
Anti-Huntingtin Protein, a.a. 181-810, clone 1HU-4C8	MAB2166
Anti-Huntingtin Protein, clone mEM48	MAB5374
Anti-Huntingtin Associated Protein 40 (HAP40)	AB5872
Polyglutamine-Expansion Diseases Marker	MAB1574
Parkinson's Disease	
Anti-Synuclein, α	AB5038P
Anti-Dopamine D2 Receptor	AB5084P
Anti-PINK1 Antibody, clone N4/49	MABN18
Anti-Tyrosine Hydroxylase	AB152
Anti-Dardarin (LRRK2)	AB9682
Anti-Parkin	AB9244
Prion Diseases	
Anti-Prion Protein	AB5058
Anti-Prion Protein, clone 2G11	MAB5542
Prion Protein	AG210
Anti-14-3-3 phospho Serine58	AB9750
Anti-Clusterin, α chain	05-354

Key Products

For a complete listing of neuroscience products from Merck Millipore, please visit **www.millipore.com/neuroguide**.

Non-Amyloid Biomarker Antibodies, Proteins, and Assays

Description	Catalogue No.
Amyotophic Lateral Sclerosis (ALS)	
Anti-Neuroketal	AB5611
Anti-Superoxide Dismutase 1 (SOD1)	AB5480
Anti-Calbindin D-28K	AB1778
Anti-p75 NTR	AB1554
Multiple Sclerosis (MS)	
Anti-Degraded Myelin	AB5864
Black-Gold 2 Myelin Marker Kit	AG105
Anti-H-CAM (CD44)	CBL1308
Anti-MOG	MAB5584
Human Oligodendrocyte Differentiation Kit (see page 9)	SCR600
General Neuronal Degeneration	
Anti-Beclin 1	AB15417
Fluoro-Jade® C	AG325
Fluoro-Ruby®	AG335
GFAP ELISA	NS830
Phosphorylated Neurofilament ELISA	NS170
ChemiKine™ Pigment Epithelium Derived Factor, Sandwich ELISA	CYT420
Mouse SAA-3 ELISA	EZMSAA3-12K
Human sICAM-1 ELISA	ECM335
Human sVCAM-1 ELISA	ECM340
LentiBrite™ GFP-LC3 lentiviral Biosensors	17-10193
LentiBrite™ GFP-LC3 Mutant lentiviral Biosensors	17-10189
LentiBrite™ GFP-PSD95 Lentiviral Biosensors	17-10227
LentiBrite™ RFP-PSD95 Lentiviral Biosensors	17-10226

Inhibitors

Description	Catalogue No.
γ-Secretase Inhibitor XX	565789
InSolution™ γ-Secretase Inhibitor X	565771
γ-Secretase Inhibitor IX	565770
γ-Secretase Inhibitor XXI, Compound E	565790
β-Secretase Inhibitor IV	565788
LRRK2-IN-1	438193
γ-Secretase Inhibitor XXIV, BMS299897	565793
β-Amyloid Oligomer Inhibitor, K01-162	200487
Bexarotene	200499

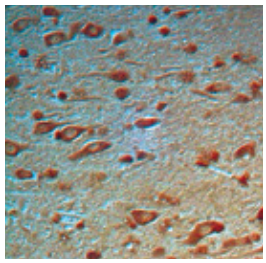
Oxidative Stress

An excess of free radicals causes oxidative stress, an unstable cellular environment that can result from exposure to alcohol, medications, poor nutrition, trauma, cold, toxins, and overexercise. Free radicals and other reactive oxygen species (ROS) form when cells encounter oxidizing agents or ionizing radiation. ROS can damage DNA, an early step in carcinogenesis; damage to other biomolecules leads to atherosclerosis, cerebral and heart ischemia-reperfusion injury, rheumatoid arthritis, inflammation, diabetes, aging, neurodegenerative diseases, and other disorders.

8-Hydroxydeoxyguanosine (8OHdG)

(Catalogue No. AB5830)

8-Hydroxydeoxyguanosine (8OHdG) is a modified base that occurs in DNA due to attack by hydroxyl radicals that are formed as byproducts and intermediates of aerobic metabolism and during oxidative stress. Merck Millipore's anti-8 hydroxyguanosine has been shown by ELISA to be completely specific for oxidized DNA while not cross-reacting with other naturally occurring nucleotides. This antibody is a valuable tool for elucidating the role of free radical damage in a number of human disease states.

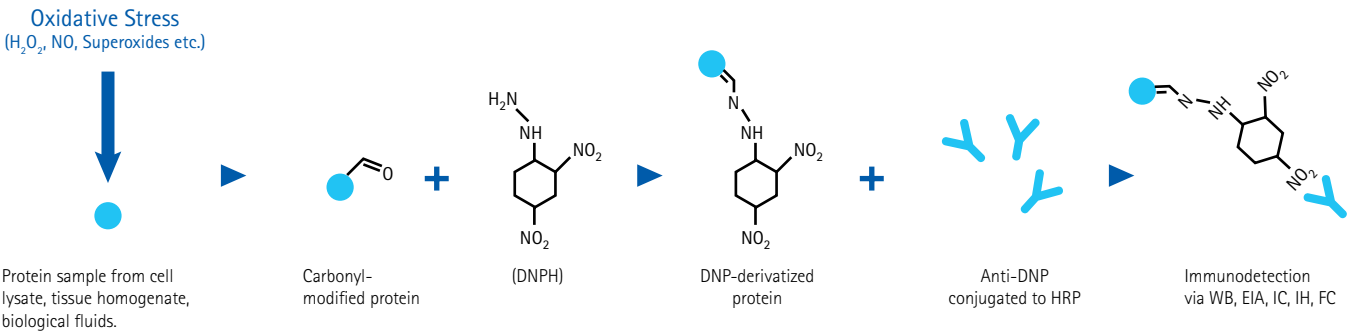


Goat anti-8-OHdG (Catalogue No. AB5830) Alzheimer's disease brain showing immunohistochemcal staining of oxidized DNA in neurons.

Technology Highlight

Oxidative stress detection with OxyBlot™, ELISA, IC, IH, & flow cytometry

Oxidative modification of proteins by oxygen free radicals and other reactive species such as hydroxynonenal occurs in physiologic and pathologic processes. As a consequence of the modification, carbonyl groups are introduced into protein side chains by a site-specific mechanism. Merck Millipore's oxidative stress detection kits enable simple and sensitive immunodetection of these carbonyl groups.

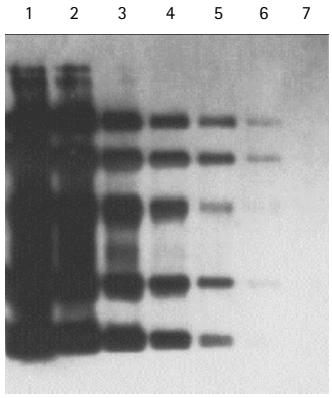


OxyBlot™ Protein Oxidation Detection Kit

(Catalogue No. S7150)

The OxyBlot™ Protein Oxidation Detection Kit provides the chemical and immunological reagents necessary to perform the immunoblot detection of carbonyl groups introduced into proteins by oxidative reactions with ozone or oxides of nitrogen or by metal catalyzed oxidation.

Under the conditions recommended in the kit, as little as 5 femtomoles of carbonyl residue can be detected. This sensitivity is at least 100 times greater than that obtained by other procedures (such as radioisotope methodology utilizing 3H-labeled NaBH4). in addition, the oxidative status of each protein can be analyzed quantitatively by comparison of the signal intensity of the same protein in different lanes on the same or different gels.

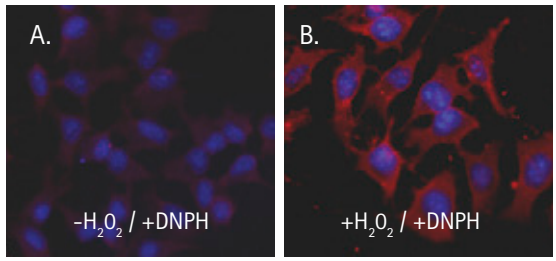


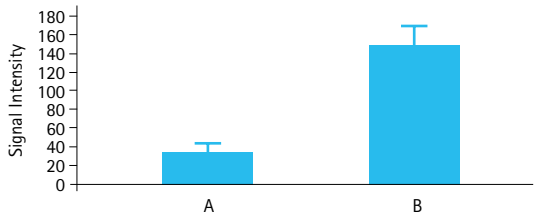
DNP in BSA Band (femtomoles)
Lane 1: 100
Lane 2: 30
Lane 3: 10
Lane 4: 3
Lane 5: 1
Lane 6: 0.3
Lane 7: 0.1

OxyICC™ Oxidized Protein Detection Kit

(Catalogue No. S7350)

Carbonyl formation is an important biomarker for oxidative stress. The OxyICC™ kit provides reagents for fluorescent immunocytochemistry of cellular protein carbonyls. This simple assay detects carbonyl modifications using dinitrophenylhydrazine (DNPH) to provide highly sensitive and quantitative results.





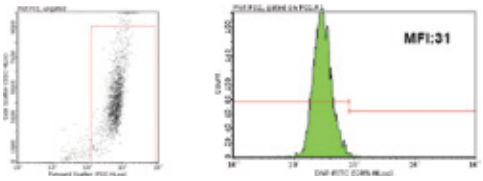
Quantitative immunodetection of protein carbonyls shows that peroxide treatment of cells results in over a four-fold signal intensity increase versus basal levels (B compared to A).

FlowCellect® Oxidative Stress Characterization Kit

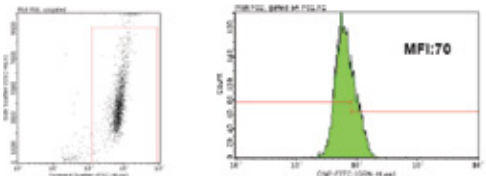
(Catalogue No. FCCH025111)

The new FlowCellect® oxidative stress characterization kit enables flow cytometric quantification of oxidative stress within cells by providing all the reagents necessary to detect carbonyl groups introduced onto proteins by reactive oxygen species. Although the assay and all of the kit components are optimized for benchtop guava easyCyte™ instruments, you can use the kit with any flow cytometer equipped with a blue (488 nm) laser.

Panel A: H_2O_2 Untreated Cells



Panel B: H_2O_2 Treated Cells



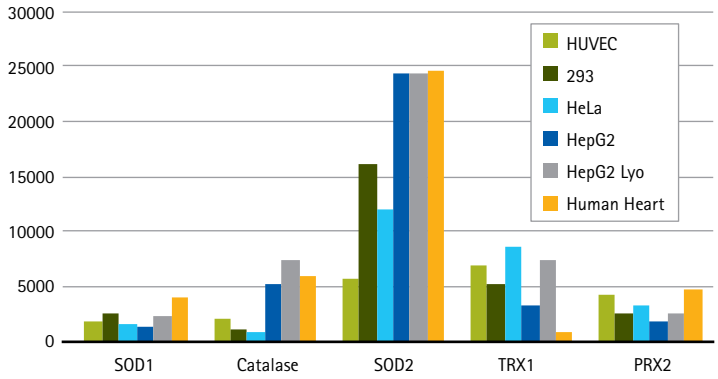
HeLa cells treated (B) with or (A) without hydrogen peroxide (H_2O_2) were processed using the FlowCellect® oxidative stress characterization kit, then analyzed on a guava easyCyte™ flow cytometer.

MILLIPLEX® MAP Human Oxidative Stress Magnetic Bead Panel

(Catalogue No. HOXSTMAG-18K)

Oxidative stress is the result of an imbalance between the production of reactive oxygen species and the removal of their reactive intermediates. ROS are products of the oxidative phosphorylation pathway. The new MILLIPLEX® MAP Human Oxidative Stress Magnetic Bead Panel enables quantitative multiplexed detection of levels of 5 oxidative stress pathway markers, including: Catalase, Peroxiredoxin 2 (PRX2), SOD1, SOD2, and Thioredoxin (TRX1).

Human Oxidative Stress Panel: Differential Expression in Cell Lines and Tissues



Multiplexed analysis of different human cell lines and heart tissue extract with the Human Oxidative Stress Magnetic Bead Panel. Lysates from different human cell lines (HUVEC, HEK 293, HeLa, HepG2) and heart tissue were prepared according to the procedures described in the protocol. 1000 ng of cell line lysates, heart tissue extract and the Lysate Control (Catalogue No. 47-231) were analyzed with the Human Oxidative Stress Magnetic Bead Panel according to the assay protocol. The Median Fluorescence Intensity (MFI) was measured with the Luminex® system.

Key Products

For a complete listing of neuroscience products from Merck Millipore, please visit **www.millipore.com/neuroguide**.

Oxidative Stress Antibodies & Inhibitors

Description	Catalogue No.
Anti-Degraded Myelin Basic Protein (MBP)	AB5864
Anti-8-Hydroxydeoxyguanosine	AB5830
Anti-Nitrotyrosine	06-284
Anti-4-Hydroxynoneal	AB5605
Anti-Neuroketal Antibody	AB5611
Anti-8-Oxoguanine Antibody, clone 483.15	MAB3560
Oxidation Inhibitor FeTMPyP	341501-10MG

Oxidative Stress Assays

Description	Catalogue No.
OxyICC™ Oxidized Protein Detection Kit	S7350
OxyBlot™ Protein Oxidation Detection Kit	S7150
OxyELISA™ Oxidized Protein Quantitation Kit	S7250
Mitochondrial Complex I Activity Assay	AAMT001
Nitrotyrosine ELISA	17-10006
OxyIH Oxidized Protein Detection Kit	S7450
MILLIPLEX® MAP Human Oxidative Phosphorylation Magnetic Bead Panel	H0XPSMAG-16K
MILLIPLEX® MAP Rat/Mouse Oxidative Phosphorylation Magnetic Bead Panel	RM0XPSMAG-17K

Neural Cell Health

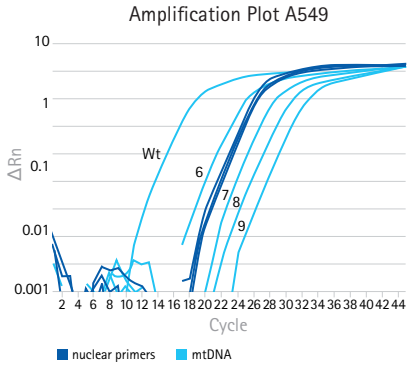
The highly intricate structure and electrical function of neurons likely account for the brain's incredibly high metabolic demands and also the ease by which drugs, environmental hazards and progressive diseases induce cellular stress and neurotoxic events. Consequent declines in neural cell health results in loss of synapses and neurons as well as reactive gliosis, all of which contribute to cognitive dysfunction and disease etiology. Merck Millipore provides a variety of unique and exceptional tools for the measure of mitochondrial and cell health, apoptosis and downstream inflammation.

Featured Product

NovaQUANT™ Human Mitochondrial to Nuclear DNA Ratio Kit

(Catalogue No. 72620-1KIT)

NovaQUANT™ quantitative real-time PCR (qPCR) assays are an innovative, reliable, and user-friendly way to determine the ratio of mitochondrial to nuclear DNA. This ratio is one measure of mitochondrial content in the cell. The accurate determination of this ratio is key to assessing cellular homeostasis, which can change with respect to cell differentiation, stress, disease, exercise, caloric intake, and toxicity.



Sensitive and linear detection of key mitochondrial and host cell genes using SYBR® Green Technology. Ethidium Bromide (EtBr)-treated A549 cells were cultured in 50 ng/mL EtBr. Passage numbers are indicated next to curves, in black. EtBr is concentrated differentially in the mitochondria due to higher mitochondrial membrane potential and subsequent DNA binding. Cells were directly lysed in PCR reactions, total DNA normalized to 1 ng/mL and targets amplified using paired mitochondrial or nuclear primers in a NovaQUANT™ qPCR assay using SYBR® Green technology. Higher passage numbers lead to a greater depletion of mtDNA as cells transition to a glycolytic energy state. Dark lines show no change in nuclear DNA. Wt equals wildtype.

Key Products

Description	Analysis	Catalogue No.
NovaQUANT™ Human Mitochondrial to Nuclear DNA Ratio Kit	qPCR assay	72620-1KIT
NovaQUANT™ Mitochondrial Biogenesis qPCR Kit	qPCR assay	72625-1KIT
P38 Stress Pathway Activation Detection Kit	Flow cytometry	FCCS025132
FlowCellect® MitoStress Kit	Flow cytometry	FCCH100109
FlowCellect® Cytochrome C Kit	Flow cytometry	FCCH100110
FlowCellect® Annexin Red Kit	Flow cytometry	FCCH100108
FlowCellect® MitoPotential Red Kit	Flow cytometry	FCCH100105
FlowCellect® MitoDamage Kit	Flow cytometry	FCCH100106
MILLIPLEX® MAP Human Late Apoptosis Magnetic Bead Panel – 3 plex	Multiplexing	48-670MAG
MILLIPLEX® MAP Human Cytokine/Chemokine Magnetic Bead Panel I	Multiplexing	HCYTOMAG-60K
MILLIPLEX® MAP Human Cytokine/Chemokine Magnetic Bead Panel II	Multiplexing	HCYP2MAG-62K
ApopTag™ Plus *In Situ* Apoptosis Fluorescein Detection Kit	Tissue staining	S7111

Detecting the Future of Neuroscience

Neuroscience is driven by difficult and insightful questions. Merck Millipore has technologies that answer. Having incorporated the expertise of Chemicon®, Upstate®, and Calbiochem®, Merck Millipore is now committed to developing comprehensive neuroscience research solutions. We are dedicated to providing quality, cutting-edge tools to the neuroscience community and look forward to a mutually beneficial collaboration for decades to come.

To Place an Order or Receive Technical Assistance

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