

#5304 Store at -20°C

PathScan® PDGFR Activity Assay: Phospho-PDGFR, Phospho-SHP2, Phospho-Akt, and Phospho-p44/42MAPK Multiplex Western Detection Cocktail II

✓ 250 µl (5 western blots)

rev. 01/29/16



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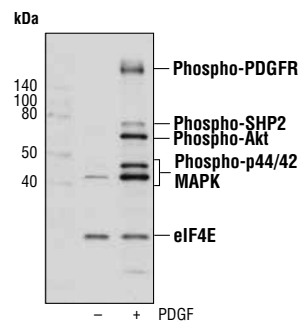
For Research Use Only. Not For Use In Diagnostic Procedures.

Applications	Species Cross-Reactivity*	Source
W Endogenous	H, M, R	Rabbit**

Antibody Cocktail Components and Molecular Weights		
No.	Antibody	Molecular Weight
4549	Phospho-PDGFR β (Tyr751) (C63G6) Rabbit mAb	190 kDa
3751	Phospho-SHP-2 (Tyr542) Antibody	72 kDa
9271	Phospho-Akt (Ser473) Antibody	60 kDa
4370	Phospho-p44/42 MAPK (Erk1/2) (Thr202/Tyr204) (D13.14.4E) Rabbit mAb	42, 44 kDa
2067	eIF4E (C46H6) Rabbit mAb	25 kDa

Description: The PathScan® Multiplex Western Detection Cocktail offers a unique method to assay the activation of multiple proteins on one membrane without stripping and reprobing. This method saves the user valuable time while increasing accuracy and minimizing reagent waste. The PDGFR Tyrosine Kinase Activity Assay allows the user to simultaneously detect the phosphorylation of PDGF receptor, SHP2, Akt and p44/42 MAPK proteins in response to PDGF. The kit also includes an eIF4E antibody to control protein loading.

Background: Platelet-derived growth factor (PDGF) is a dimeric molecule that exists as homodimers or heterodimers of related polypeptide chains (A and B). Two types of PDGF receptors have been identified. The PDGF alpha-receptor binds all three isoforms with high affinity, whereas the beta-receptor binds only PDGF-BB with high affinity, PDGF-AB with low affinity and does not appear to bind PDGF-AA (1). PDGF exerts its stimulatory effects on cells by binding to these two related protein tyrosine kinase receptors. Ligand binding induces receptor dimerization and autophosphorylation, allowing binding and activation of cytoplasmic SH2-domain-containing signal transduction molecules. Thereby, a number of different signaling pathways are initiated, leading to cell growth, actin reorganization, migration and differentiation (2-4). In clinical studies, PDGF expression has been shown in a number of different solid tumors, from glioblastomas to prostate carcinomas. In these various tumor types, the biologic role of PDGF signaling can vary from autocrine stimulation of cancer cell growth to more subtle paracrine interactions involving adjacent stroma and even angiogenesis. Targeting PDGF signaling becomes an effective way for tumor treatment (5).



Western blot analysis of extracts from serum-starved NIH/3T3 cells untreated or PDGF-treated (#8912, 50 ng/ml for 20 minutes), using PathScan PDGFR Receptor Activity Assay cocktail to detect phosphorylation of PDGFR, SHP2, Akt and p44/42 MAPK.

Specificity/Sensitivity: Each phospho-antibody in this cocktail recognizes endogenous levels of only the phosphorylated form of its specific target. The eIF4E antibody detects endogenous levels of its target independent of phosphorylation and is provided to control for protein loading.

Source/Purification: Antibodies are produced by immunizing animals with synthetic peptides, and are purified by combinations of Protein A and peptide affinity chromatography.

Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA and 50% glycerol. Store at -20°C. Do not aliquot the antibody.

*Species cross-reactivity is determined by western blot, using the individual antibody cocktail components.

**Anti-rabbit secondary antibodies must be used to detect this antibody cocktail.

Recommended Antibody Dilutions:
Western blotting 1:200

For application specific protocols please see the web page for this product at www.cellsignal.com.

Please visit www.cellsignal.com for a complete listing of recommended companion products.

Background References:

- (1) Westermark, B. et al. (1990) *Ciba Found. Symp.* 150, 6–22.
- (2) Heldin, C.H. (1997) *FEBS Lett.* 410, 17–21.
- (3) Bornfeldt, K. E. et al. (1995) *Ann. N. Y. Acad. Sci.* 766, 416–430.
- (4) Renhowe, P.A. (2002) *Curr. Opin. Drug Discov. Devel.* 5, 214–224.
- (5) George, D. (2001) *Semin. Oncol.* 28, 27–33.v

IMPORTANT: For Western blots, incubate membrane with diluted antibody in 5% w/v BSA, 1X TBS, 0.1% Tween-20 at 4°C with gentle shaking, overnight.

Rabbit monoclonal antibody is produced under license (granting certain rights including those under U. S. Patent No. 5,675,063) from Epitomics, Inc.

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Applications Key: W—Western IP—Immunoprecipitation IHC—Immunohistochemistry IC—Immunocytochemistry IF—Immunofluorescence F—Flow cytometry E—ELISA D—DELFA®
Species Cross-Reactivity Key: H—human M—mouse R—rat Hm—hamster Mk—monkey Mi—mink C—chicken X—Xenopus Z—zebra fish B—bovine All—all species expected
Species enclosed in parentheses are predicted to react based on 100% sequence homology.

Material Safety Data Sheet (MSDS) for Antibodies

rev. 08/09/07

I. Identification:

Product name: Antibodies

Product Catalog Number: Includes antibodies within the following range of catalog numbers: 2000-5999, 7000-7999 and 9000-9999.

CAS number: None

Manufacturer Supplier: Cell Signaling Technology
3 Trask Lane
Danvers, MA 01923 USA
1-978-867-2300 TEL
1-978-867-2400 FAX
1-978-578-6737 Emergency Phone

II. Composition/Information on Ingredients:

This product is composed of antibodies in aqueous buffer solution. According to 29 CFR 1910.1200(d), hazardous ingredients at less than <1% and carcinogens at less than <0.1% are considered non-hazardous. Any hazardous or carcinogenic ingredients exceeding these criteria are listed below.

This product may contain the following hazardous ingredients.

Ingredient	CAS#	Percent
Glycerol	56-81-5	50%

III. Hazard Identification:

Emergency Overview of Hazardous ingredient: Glycerol (CAS# 56-81-5)

Caution: Avoid contact and inhalation.

Target Organ: Kidneys.

NFPA Rating:

Health Rating:	1
Flammability Rating:	0
Reactivity Rating:	0

IV. First Aid Measures:

Inhalation: If inhaled, remove to fresh air. If breathing is difficult, get medical attention.

Ingestion: If swallowed and person is conscious, rinse out mouth with water. Get medical attention.

Skin Exposure: In case of contact, wash skin with soap and water.

Eye Exposure: In case of contact with eyes, immediately flush eyes water for at least 15 minutes. Get medical attention.

V. Fire Fighting Measures:

Flash Point: Data not available.

Autoignition Temperature: Data not available.

Fire Extinguishing Media: Water spray, dry chemical, foam, or carbon dioxide.

Firefighting: Wear protective clothing and self-contained breathing apparatus to prevent contact with skin and eyes.

VI. Accidental Release Measures:

Absorb liquid with an absorbent material. Transfer contaminated absorbent to a chemical waste container for disposal.

VII. Handling And Storage:

Avoid inhalation and contact with eyes and skin. Avoid prolonged or repeated exposure.

Store at -20°C in tightly closed container.

VIII. Exposure Controls/Personal

Engineering Controls: Maintain adequate ventilation, eye wash and quick-drench facilities in work area.

Personal Protective Equipment: Lab coat, chemical resistant gloves and chemical safety glasses.

Occupational Exposure Limits: Data not available.

IX. Exposure Controls/Personal Protection:

Physical State:	Colorless liquid.
Odor:	Odorless.
Boiling Point:	Data not available.
Melting Point:	Data not available.
Hazardous Organic Compound:	Data not available.
Solubility in water:	Readily miscible in water.

X. Stability and Reactivity:

Stability: Stable.

Hazardous Decomposition: May form carbon dioxide and carbon monoxide.

Conditions to avoid: Strong oxidizing agents

XI. Toxicological Information:

May cause skin irritation.

May be toxic if absorbed through skin or ingested.

May cause eye irritation.

Target Organs: Kidneys

Prolonged exposure may cause nausea, headache, and vomiting.

XII. Ecological Information:

Data not available.

XIII. Disposal Considerations:

Dispose of in accordance with federal, state and local environmental regulations.

XIV. Transport Information:

D.O.T.: This substance is considered non-hazardous for transport.

IATA: This substance is considered non-hazardous for air transport.

XV. Regulatory Information:

EU Regulation/Classification/Labeling Information: Not available for this product.

Chemical Inventory Status:

SARA Listed Component: None.

TSCA Listed Component: None.

Canada (WHMIS): DSL No, NDSL No.

XVI. Other Information:

This compound is sold only for research use by personnel familiar with chemicals and who are well trained in good laboratory habits, such as avoiding spills, keeping hands clean at all times and not rubbing eyes with hands while working in the laboratory.

This solution is sold only in microliter quantities for use in life sciences research. No other use is intended, and any other use may involve substantive hazards.

The above information is believed to be correct but does not purport to be all inclusive and shall be used only as a guide for experienced personnel. Cell Signaling Technology, Inc., shall not be held liable for any damage resulting from the handling of or from contact with the above product. The burden of safe use of this material rests entirely with the user.