

### DESCRIPTION

<b>Species Reactivity</b>	Human/Mouse
<b>Specificity</b>	Detects human and mouse SHP-2 when phosphorylated at Y542.
<b>Source</b>	Polyclonal Rabbit IgG
<b>Purification</b>	Antigen Affinity-purified
<b>Immunogen</b>	Phosphopeptide containing human SHP-2 Y542 site
<b>Formulation</b>	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS.

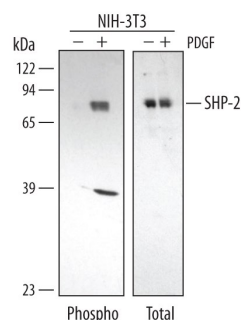
### APPLICATIONS

**Please Note:** Optimal dilutions should be determined by each laboratory for each application. *General Protocols* are available in the Technical Information section on our website.

	Recommended Concentration	Sample
<b>Western Blot</b>	1 µg/mL	See Below
<b>Immunocytochemistry</b>	3-15 µg/mL	Immersion fixed THP-1 human acute monocytic leukemia cell line treated with PMA
<b>Simple Western</b>	10 µg/mL	See Below

### DATA

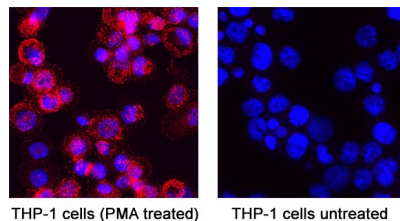
#### Western Blot



#### Detection of Mouse Phospho-SHP-2 (Y542) by Western Blot.

Western blot shows lysates of NIH-3T3 mouse embryonic fibroblast cell line untreated (-) or treated (+) with 50 ng/mL Human PDGF-BB (Catalog # [Catalog # 220-BB](#)) for 20 minutes. PVDF membrane was probed with 1 µg/mL of Rabbit Anti-Human/Mouse Phospho-SHP-2 (Y542) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3790), followed by HRP-conjugated Anti-Rabbit IgG Secondary Antibody (Catalog # [Catalog # HAF008](#)). A specific band was detected for Phospho-SHP-2 (Y542) at approximately 72 kDa (as indicated). The lysates were also probed for total SHP-2 with Human/Mouse/Rat SHP-2 Antigen Affinity-purified Polyclonal Antibody (Catalog # [Catalog # AF1894](#)). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

#### Immunocytochemistry

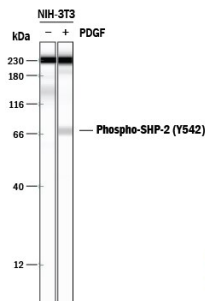


THP-1 cells (PMA treated)

THP-1 cells untreated

**SHP-2 in THP-1 Human Cell Line.** SHP-2 was detected in immersion fixed THP-1 human acute monocytic leukemia cell line treated with PMA (positive staining) and THP-1 human acute monocytic leukemia cell line (untreated; negative staining) using Rabbit Anti-Human/Mouse Phospho-SHP-2 (Y542) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3790) at 3 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Rabbit IgG Secondary Antibody (red; Catalog # [NL004](#)) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm. Staining was performed using our protocol for Fluorescent ICC Staining of Non-adherent Cells.

## Simple Western



### Detection of Mouse Phospho-SHP-2 (Y542) by Simple Western™

Simple Western™ lane view shows lysates of NIH-3T3 mouse embryonic fibroblast cell line untreated (-) or treated (+) with 50 ng/mL Recombinant Human PDGF-BB (Catalog # 220-BB) for 20 minutes, loaded at 0.2 mg/mL. A specific band was detected for Phospho-SHP-2 (Y542) at approximately 72 kDa (as indicated) using 10 µg/mL of Rabbit Anti-Human/Mouse Phospho-SHP-2 (Y542) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3790). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.\*Non-specific interaction with the 230 kDa Simple Western standard may be seen with this antibody.

## PREPARATION AND STORAGE

<b>Reconstitution</b>	Reconstitute at 0.2 mg/mL in sterile PBS.
<b>Shipping</b>	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
<b>Stability &amp; Storage</b>	<p><b>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</b></p> <ul style="list-style-type: none"> <li>• 12 months from date of receipt, -20 to -70 °C as supplied.</li> <li>• 1 month, 2 to 8 °C under sterile conditions after reconstitution.</li> <li>• 6 months, -20 to -70 °C under sterile conditions after reconstitution.</li> </ul>

## BACKGROUND

Src-Homology domain-2 containing protein tyrosine Phosphatase 2 (SHP-2), also called protein tyrosine phosphatase, non-receptor type 11 (PTPN11), PTP1D, PTP2C, and SYP, is an enzyme that dephosphorylates tyrosine residues in proteins. The protein contains two Src homology 2 (SH2) domains, which both regulate the activity of the enzyme (1) and allow it to selectively bind to SH2 sites on proteins such as Dok1, IRS1, and the insulin receptor (2). SHP-2 plays a unique stimulatory role in cell signaling. Cells lacking SHP-2 have poor mobility because the hyper-phosphorylation of FAK and other proteins in the focal adhesion complex (3) prevents turnover of cellular attachment points. Without SHP-2, sustained ERK stimulation does not take place (4). The Y992 phosphorylation site of EGFR is a particularly good substrate for SHP-2 (5) and a phosphopeptide containing this sequence can be used to measure the activity of the enzyme (R&D Systems, Catalog # ES006) by detecting release of phosphate (R&D Systems, Catalog # DY996).

### References:

1. Zhao, Z. *et al.* (1994) J. Biol. Chem. **269**:8780.
2. Clemmons, D.R. and Maile, L.A. (2005) Mol. Endocrinol. **19**:1.
3. von Wichert, G. *et al.* (2003) EMBO J. **22**:5023.
4. Maroun, C.R. *et al.* (2000) Mol. Cell. Biol. **20**:8513.
5. Sugimoto, S. *et al.* (1993) J. Biol. Chem. **269**:22771.