

DESCRIPTION	
<b>Species Reactivity</b>	Mouse/Rat
<b>Specificity</b>	Detects mouse and rat MyD88 in Western blots.
<b>Source</b>	Polyclonal Goat IgG
<b>Purification</b>	Antigen Affinity-purified
<b>Immunogen</b>	<i>E. coli</i> -derived recombinant mouse MyD88 Met1-Pro296 Accession # Q3U7M4
<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>	5-15 µg/mL	See Below
<b>Intracellular Staining by Flow Cytometry</b>	2.5 µg/10 <sup>6</sup> cells	See Below
<b>Simple Western</b>	10 µg/mL	See Below
<b>CyTOF-ready</b>	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.	

**DATA**

**Western Blot**

**Detection of Mouse/Rat MyD88 by Western Blot.** Western blot shows lysates of L6 rat myoblast cell line, A20 mouse B cell lymphoma cell line, and CH-1 mouse B cell lymphoma cell line. PVDF membrane was probed with 1 µg/mL of Goat Anti-Mouse/Rat MyD88 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3109) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF017). A specific band was detected for MyD88 at approximately 39 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 2.

**Immunocytochemistry**

**MyD88 in RAW 264.7 Mouse Cell Line.** MyD88 was detected in immersion fixed RAW 264.7 mouse monocyte/macrophage cell line using Goat Anti-Mouse/Rat MyD88 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3109) at 1.7 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Goat IgG Secondary Antibody (red; Catalog # NL001) and counterstained with DAPI (blue). Specific staining was localized to cytoplasmic. View our protocol for Fluorescent ICC Staining of Cells on Coverslips.

**Simple Western**

**Detection of Rat MyD88 by Simple Western™.** Simple Western lane view shows lysates of L6 rat myoblast cell line, loaded at 0.2 mg/mL. A specific band was detected for MyD88 at approximately 39 kDa (as indicated) using 10 µg/mL of Goat Anti-Mouse/Rat MyD88 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3109) followed by 1:50 dilution of HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF109). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.

**Intracellular Staining by Flow Cytometry**

**Detection of MyD88 in Mouse Splenocytes by Flow Cytometry.** Mouse splenocytes were stained with Goat Anti-Mouse/Rat MyD88 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3109, filled histogram) or control antibody (Catalog # AB-108-C, open histogram), followed by Allophycocyanin-conjugated Anti-Goat IgG Secondary Antibody (Catalog # F0108). To facilitate intracellular staining, cells were fixed with paraformaldehyde and permeabilized with saponin. This application has not been tested in rat samples.

**PREPARATION AND STORAGE**

**Reconstitution** Reconstitute at 0.2 mg/mL in sterile PBS.

**Shipping** 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

**Stability & Storage** Use a manual defrost freezer and avoid repeated freeze-thaw cycles.

- 12 months from date of receipt, -20 to -70 °C as supplied.
- 1 month, 2 to 8 °C under sterile conditions after reconstitution.
- 6 months, -20 to -70 °C under sterile conditions after reconstitution.

**BACKGROUND**

Myeloid Differentiation primary response protein 88 (MyD88) is a 296 amino acid, 34 kDa, ubiquitously expressed, cytoplasmic adaptor protein involved in the signaling of TLR and IL-1 R family members. MyD88 contains an N-terminal death domain and a C-terminal Toll/IL-1 R (TIR) domain. Each domain seems to participate in protein-protein interactions, as the death domain is inactive. Upon Toll receptor ligation, MyD88 is recruited to the receptor and initiates a signaling cascade that results in NkκB and JNK activation. The amino acid sequence of mouse MyD88 is 93% identical to that of rat MyD88.