

**DESCRIPTION**

<b>Species Reactivity</b>	Mouse
<b>Specificity</b>	Detects mouse Osteopontin /OPN in direct ELISAs.
<b>Source</b>	Recombinant Monoclonal Rabbit IgG Clone # 2139B
<b>Purification</b>	Protein A or G purified from cell culture supernatant
<b>Immunogen</b>	Mouse myeloma cell line NS0-derived recombinant mouse Osteopontin/OPN Leu17-Asn294 Accession # Q547B5
<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.

	<b>Recommended Concentration</b>	<b>Sample</b>
<b>Western Blot</b>	1 µg/mL	See Below
<b>Immunocytochemistry</b>	3-25 µg/mL	See Below
<b>Immunohistochemistry</b>	3-25 µg/mL	See Below

**DATA**

**Western Blot**

**Detection of Mouse Osteopontin/OPN by Western Blot.** Western blot shows conditioned media from Neuro-2A mouse neuroblastoma cell line (negative control) and L-929 mouse fibroblast cell line. PVDF membrane was probed with 1 µg/mL of Rabbit Anti-Mouse Osteopontin/OPN Monoclonal Antibody (Catalog # MAB808) followed by HRP-conjugated Anti-Rabbit IgG Secondary Antibody (Catalog # HAF008). A specific band was detected for Osteopontin/OPN at approximately 50 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

**Immunocytochemistry**

**Osteopontin/OPN in RAW 264.7 Mouse Cell Line.** Osteopontin/OPN was detected in immersion fixed RAW 264.7 mouse monocyte/macrophage cell line using Rabbit Anti-Mouse Osteopontin/OPN Monoclonal Antibody (Catalog # MAB808) 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. View our protocol for [Fluorescent ICC Staining of Non-adherent Cells](#).

**Immunohistochemistry**

**Osteopontin/OPN in Mouse Kidney.** Osteopontin/OPN was detected in perfusion fixed frozen sections of mouse kidney using Rabbit Anti-Mouse Osteopontin/OPN Monoclonal Antibody (Catalog # MAB808) at 3 µg/mL for 1 hour at room temperature followed by incubation with the Anti-Rabbit IgG VisUCyte™ HRP Polymer Antibody (Catalog # VC003). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Specific staining was localized to convoluted tubules. View our protocol for [IHC Staining with VisUCyte HRP Polymer Detection Reagents](#).

**PREPARATION AND STORAGE**

<b>Reconstitution</b>	Reconstitute at 0.5 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>	<b>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</b> <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**

Osteopontin (OPN), previously called SPP1 (secreted phosphoprotein 1), Eta-1 (early T lymphocyte activation 1) or BSP (bone sialoprotein), is a secreted molecule in the SIBLING (small integrin-binding ligand N-linked glycoprotein) family of non-collagenous matricellular proteins (1-3). Mouse OPN is synthesized as a 294 amino acid (aa) precursor protein with a 16 aa signal peptide and a 278 aa mature protein (3). Mature mouse OPN shares 79% and 64% aa sequence identity with rat and human OPN, respectively. OPN is highly acidic and has 26 potential Ser/Thr phosphorylation sites and a C-terminal CD44 binding site (1-4). Depending on tissue-specific modification by O- and N-glycosylation, sulfation, phosphorylation and transglutamination, OPN can be detected at 45-75 kDa (5, 6). The central region of OPN contains RGD and non-RGD binding sites for multiple integrins (3, 4). Adjacent to the RGD motif is the sequence SLAYGLR (SVVYGLR in human) which serves as a cryptic binding site for additional integrins: it is masked in full length OPN but is exposed following OPN cleavage by thrombin in tumors and sites of tissue injury (6-8). OPN can also be cleaved by MMP-3, -7, -9, and -12 within the SLAYGLR motif and at sites closer to the C-terminus (8, 9). OPN is widely expressed and is prominent in mineralized tissues. It inhibits bone mineralization and kidney stone formation, and promotes inflammation and cell adhesion and migration (1, 2, 4, 6). Its expression is up-regulated during inflammation, obesity, atherosclerosis, cancer, and tissue damage, and contributes to the pathophysiology of these conditions (1, 2, 6, 9, 10).

**References:**

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