

## DESCRIPTION

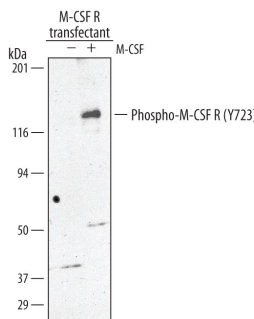
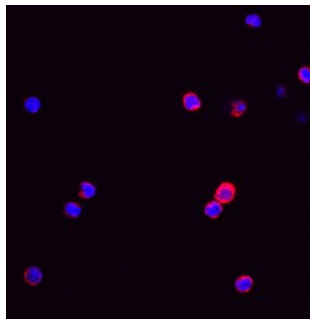
<b>Species Reactivity</b>	Human
<b>Specificity</b>	Detects human M-CSF R when phosphorylated at Y723 in Western blots.
<b>Source</b>	Polyclonal Rabbit IgG
<b>Purification</b>	Antigen and protein A Affinity-purified
<b>Immunogen</b>	Phosphopeptide containing human M-CSF R Y723 site
<b>Formulation</b>	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied 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>	0.5 µg/mL	See Below
<b>Immunocytochemistry</b>	5-15 µg/mL	See Below

## DATA

<b>Western Blot</b>	<b>Immunocytochemistry</b>
 <p><b>Detection of Human Phospho-M-CSF R (Y723) by Western Blot.</b> Western blot shows lysates of M-CSF R-transfected NSO mouse myeloma cell line untreated (-) or treated (+) with 100 ng/mL Recombinant Human M-CSF (Catalog # 216-MC) for 10 minutes. PVDF membrane was probed with 0.5 µg/mL of Rabbit Anti-Human Phospho-M-CSF R (Y723) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3268), followed by HRP-conjugated Anti-Rabbit IgG Secondary Antibody (Catalog # HAF008). A specific band was detected for Phospho-M-CSF R (Y723) at approximately 175 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.</p>	 <p><b>M-CSF R in Human PBMCs.</b> M-CSF R phosphorylated at Y723 was detected in immersion fixed human peripheral blood mononuclear cells (PBMCs) using Rabbit Anti-Human Phospho-M-CSF R (Y723) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3268) at 10 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). View our protocol for <a href="#">Fluorescent ICC Staining of Non-adherent Cells</a>.</p>

## 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

M-CSF R, the product of the *c-fms* proto-oncogene, is a member of the type III subfamily of receptor tyrosine kinases that also includes receptors for SCF and PDGF. These receptors each contain five immunoglobulin-like domains in their extracellular domain (ECD) and a split kinase domain in their intracellular region (1-4). M-CSF R is expressed primarily on cells of the monocyte/macrophage lineage, dendritic cells, stem cells and in the developing placenta (1). Human M-CSF R cDNA encodes a 972 amino acid (aa) type I membrane protein with a 19 aa signal peptide, a 493 aa extracellular region containing the ligand-binding domain, a 25 aa transmembrane domain and a 435 aa cytoplasmic domain. The human M-CSF R ECD shares 60%, 64%, 72%, 75%, 75% and 76% aa identity with mouse, rat, bovine, canine, feline and equine M-CSF R, respectively. Activators of protein kinase C induce TACE/ADAM17 cleavage of the M-CSF R, releasing the functional ligand-binding extracellular domain (5). M-CSF binding induces receptor homodimerization, resulting in transphosphorylation of specific cytoplasmic tyrosine residues and signal transduction (6). The intracellular domain of activated M-CSF R binds more than 150 proteins that affect cell proliferation, survival, differentiation and cytoskeletal reorganization. Among these, PI3Kinase, P42/44 ERK and c-Cbl are key transducers of M-CSF R signals (3, 4). M-CSF R engagement is continuously required for macrophage survival and regulates lineage decisions and maturation of monocytes, macrophages, osteoclasts and DC (3, 4). M-CSF R and integrin  $\alpha\beta_3$  share signaling pathways during osteoclastogenesis and deletion of either causes osteopetrosis (7, 8). In the brain, microglia expressing increased M-CSF R are concentrated with Alzheimers  $\alpha\beta$  peptide, but their role in pathogenesis is unclear (9, 10).

## References:

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