

#### DESCRIPTION

<b>Species Reactivity</b>	Human
<b>Specificity</b>	Detects human TNF- $\alpha$ in direct ELISAs.
<b>Source</b>	Monoclonal Human IgG <sub>1</sub> Clone # 6402
<b>Purification</b>	Protein A or G purified from cell culture supernatant
<b>Immunogen</b>	<i>E. coli</i> -derived recombinant human TNF- $\alpha$ Gly57-Leu233 Accession # P01375
<b>Conjugate</b>	Alexa Fluor 647 Excitation Wavelength: 650 nm Emission Wavelength: 668 nm
<b>Formulation</b>	Supplied 0.2 mg/mL in a saline solution containing BSA and Sodium Azide  *Contains <0.1% Sodium Azide, which is not hazardous at this concentration according to GHS classifications. Refer to the Safety Data Sheet (SDS) for additional information and handling instructions.

#### 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>Intracellular Staining by Flow Cytometry</b>	0.25-1 $\mu$ g/10 <sup>6</sup> cells	Human peripheral blood mononuclear cells (PBMCs) treated with PMA and Ca <sup>2+</sup> ionomycin fixed with Flow Cytometry Fixation Buffer (Catalog # FC004) and permeabilized with Flow Cytometry Permeabilization/Wash Buffer I (Catalog # FC005)

#### PREPARATION AND STORAGE

<b>Shipping</b>	The product is shipped with polar packs. Upon receipt, store it immediately at the temperature recommended below.
<b>Stability &amp; Storage</b>	<b>Protect from light. Do not freeze.</b> <ul style="list-style-type: none"> <li>12 months from date of receipt, 2 to 8 °C as supplied.</li> </ul>

#### BACKGROUND

Tumor necrosis factor alpha (TNF-alpha, TNF- $\alpha$ , TNFA), also known as Cachectin and TNFSF2, is the prototypic ligand of the TNF superfamily. It is a pleiotropic molecule that plays a central role in inflammation, immune system development, apoptosis, and lipid metabolism. TNF- $\alpha$  is produced by several lymphoid cells as well as by astrocytes, endothelial cells, and smooth muscle cells. Human TNF-alpha consists of a 35 amino acid (aa) cytoplasmic domain, a 21 aa transmembrane segment, and a 177 aa extracellular domain (ECD). Within the ECD, human TNF-alpha shares 97% aa sequence identity with rhesus and 71%-92% with bovine, canine, cotton rat, equine, feline, mouse, porcine, and rat TNF-alpha. TNF-alpha is produced by a wide variety of immune, epithelial, endothelial, and tumor cells. TNF-alpha is assembled intracellularly to form a noncovalently linked homotrimer which is expressed on the cell surface. Cell surface TNF-alpha can induce the lysis of neighboring tumor cells and virus infected cells, and it can generate its own downstream cell signaling following ligation by soluble TNFR I. Shedding of membrane bound TNF-alpha by TACE/ADAM17 releases the bioactive cytokine, a 55 kDa molecular weight soluble trimer of the TNF-alpha extracellular domain. TNF-alpha binds the ubiquitous 55-60 kDa TNF RI and the hematopoietic cell-restricted 80 kDa TNF RII, both of which are also expressed as homotrimers present on virtually all cell types. Both type I and type II receptors bind TNF-alpha with comparable affinity, although only TNF RI contains a cytoplasmic death domain which triggers the activation of apoptosis. Soluble forms of both types of receptors are released and can neutralize the biological activity of TNF-alpha.

#### PRODUCT SPECIFIC NOTICES

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