

Product Datasheet

TNF alpha Rabbit Polyclonal Antibody (orb526980)

Catalog Number	orb526980
Category	Antibodies
Description	TNF alpha Rabbit Polyclonal Antibody
Target	Non-structural protein 3
Clonality	Polyclonal
Species/Host	Rabbit
Isotype	Rabbit IgG
Conjugation	Unconjugated
Reactivity	Human, Mouse, Rat
Form/Appearance	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 µg/ml.
Buffer/Preservatives	Each vial contains 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na ₂ HPO ₄ .
Reconstitution	Add 0.2ml of distilled water will yield a concentration of 500ug/ml.
Purification	Immunogen affinity purified.
Immunogen	A synthetic peptide corresponding to a sequence at the C-terminus of human TNF alpha, different from the related mouse sequence by five amino acids, and rat sequence by seven amino acids.
UniProt ID	P01375
MW	25 kDa

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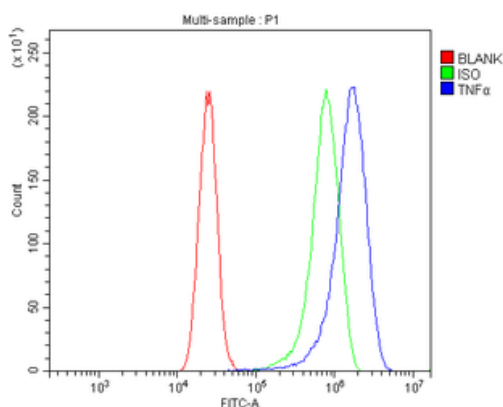
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Tested applications	FC, IHC, WB
Dilution range	Western blot, 0.1-0.5µg/ml, Human, Mouse, Rat Immunohistochemistry (Paraffin-embedded Section), 2-5µg/ml, Human Flow Cytometry (Fixed), 1-3 µg/1x10 ⁶ cells, Human
Specificity	No cross reactivity with other proteins.
Cross Reactivity	No cross-reactivity with other proteins
Antibody Type	Primary Antibody
Storage	Maintain refrigerated at 2-8°C for up to 2 weeks. For long term storage store at -20°C in small aliquots to prevent freeze-thaw cycles.
Note	For research use only
Expiration Date	12 months from date of receipt.



Flow Cytometry analysis of CACO-2 cells using anti-TNF alpha antibody. Overlay histogram showing CACO-2 cells (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-TNF alpha Antibody (1 µg/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (5-10 µg/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1 µg/1x10⁶) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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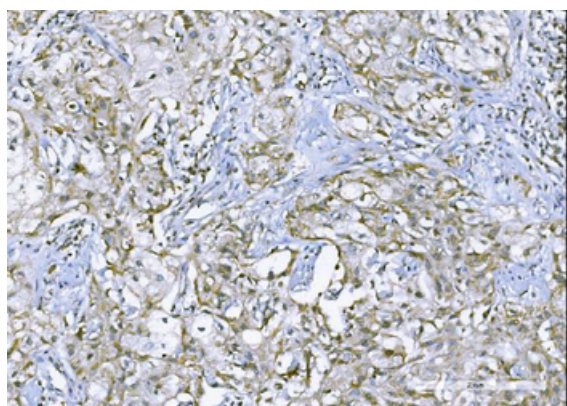
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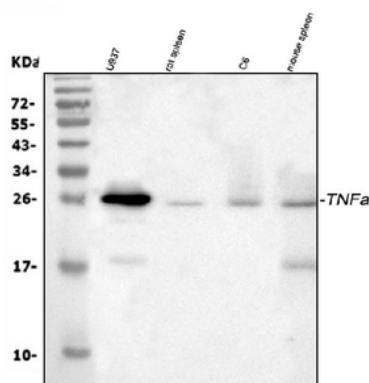
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IHC analysis of TNF alpha using anti-TNF alpha antibody. TNF alpha was detected in a paraffin-embedded section of human B lymphocytic tumor tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml rabbit anti-TNF alpha Antibody overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit with DAB as the chromogen.



Western blot analysis of TNF alpha using anti-TNF alpha antibody. Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 µg of sample under reducing conditions. Lane 1: human U937 whole cell lysates, Lane 2: rat spleen tissue lysates, Lane 3: rat C6 whole cell lysates, Lane 4: mouse spleen tissue lysates. After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-TNF alpha antigen affinity purified polyclonal antibody at 0.5 µg/mL overnight at 4°C, then washed with TBS-0.1% Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:5000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit with Tanon 5200 system. A specific band was detected for TNF alpha at approximately 25 kDa. The expected band size for TNF alpha is at 26 kDa.

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