

Recombinant human TNF alpha protein ab9642

20 References [画像数 7](#)

製品の詳細

製品名	Recombinant human TNF alpha protein		
生理活性	The ED ₅₀ , as determined by the cytolysis of murine L929 cells in the presence of Actinomycin D, is ≤ 0.05 ng/mL, corresponding to a specific activity of ≥ 2 x 10 ⁷ units/mg.		
精製度	> 98 % SDS-PAGE. >98% by HPLC analyses. Sterile filtered.		
エンドトキシン・レベル	< 1.000 Eu/μg		
発現系	Escherichia coli		
アクセッション番号	<u>P01375</u>		
タンパク質長	Full length protein		
Animal free	No		
由来	Recombinant		
生物種	Human		
配列	VRSSRTPSD KPVAVVANP QAEGQLQWLN RRANALLANG VELRDNQLVV PSEGLYLIYS QVLFKGGQCP STHVLLTHTI SRIAVSYQTK VNLLSAIKSP CQRETPEGAE AKPWYEPIYL GGVFQLEKGD RLSAEINRPD YLDFAESGQV YFGIIAL		
予測される分子量	17 kDa		
領域	77 to 233		
配列の追加情報	aa 77 to 233 refers to the full length mature form (soluble).		

特性

Our **Abpromise guarantee** covers the use of **ab9642** in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

アプリケーション	Functional Studies
	Sandwich ELISA
	HPLC
	SDS-PAGE
製品の状態	Lyophilized

備考 Lots prior to June 2015 contain 0.036% Tris (including lots GR157466-11, -12 and -13).

前処理および保存

保存方法および安定性 Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -80°C. Avoid freeze / thaw cycle.

Constituents: 0.049% Sodium phosphate, 0.12% Sodium chloride

This product is an active protein and may elicit a biological response in vivo, handle with caution.

再構成 Reconstitute with dH2O to make a final concentration between 0.1 to 1.0 mg/ml.

関連情報

機能 Cytokine that binds to TNFRSF1A/TNFR1 and TNFRSF1B/TNFR. It is mainly secreted by macrophages and can induce cell death of certain tumor cell lines. It is potent pyrogen causing fever by direct action or by stimulation of interleukin-1 secretion and is implicated in the induction of cachexia, Under certain conditions it can stimulate cell proliferation and induce cell differentiation.

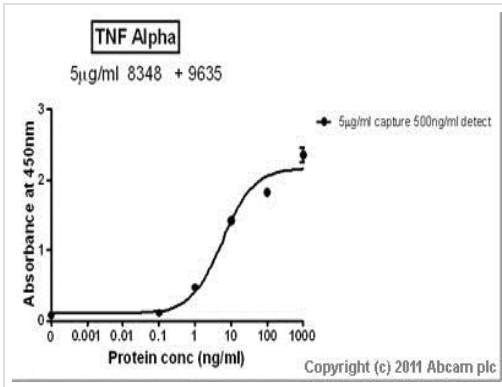
関連疾患 Genetic variations in TNF are a cause of susceptibility psoriatic arthritis (PSORAS) [MIM:607507]. PSORAS is an inflammatory, seronegative arthritis associated with psoriasis. It is a heterogeneous disorder ranging from a mild, non-destructive disease to a severe, progressive, erosive arthropathy. Five types of psoriatic arthritis have been defined: asymmetrical oligoarthritis characterized by primary involvement of the small joints of the fingers or toes; asymmetrical arthritis which involves the joints of the extremities; symmetrical polyarthritis characterized by a rheumatoidlike pattern that can involve hands, wrists, ankles, and feet; arthritis mutilans, which is a rare but deforming and destructive condition; arthritis of the sacroiliac joints and spine (psoriatic spondylitis).

配列類似性 Belongs to the tumor necrosis factor family.

翻訳後修飾 The soluble form derives from the membrane form by proteolytic processing. The membrane form, but not the soluble form, is phosphorylated on serine residues. Dephosphorylation of the membrane form occurs by binding to soluble TNFRSF1A/TNFR1. O-glycosylated; glycans contain galactose, N-acetylgalactosamine and N-acetylneuraminic acid.

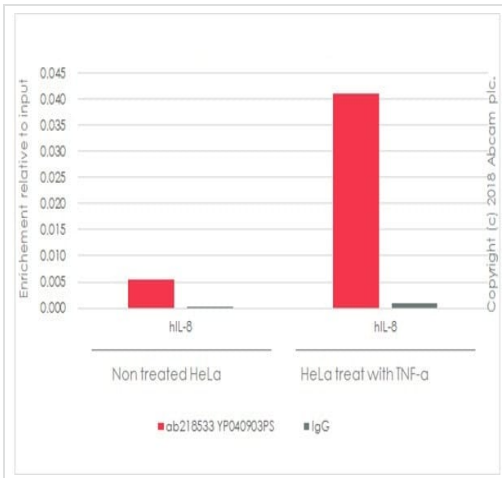
細胞内局在 Secreted and Cell membrane.

画像



Sandwich ELISA - Recombinant human TNF alpha protein (ab9642)

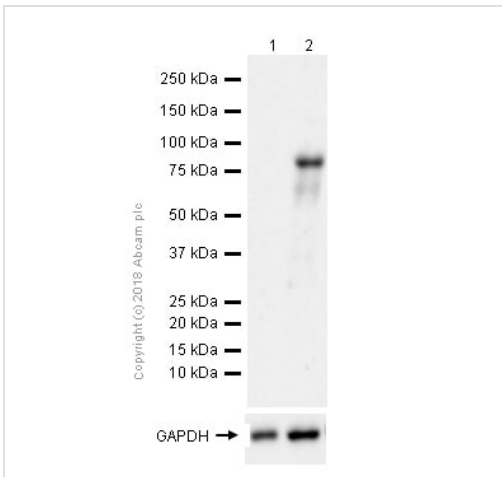
Standard curve for TNF alpha (Analyte: ab9642); dilution range 1µg/ml to 1µg/ml using Capture Antibody Mouse monoclonal [2C8] to TNF alpha ([ab8348](#)) at 5µg/ml and Detector Antibody Rabbit polyclonal to TNF alpha ([ab9635](#)) at 0.5µg/ml.



ChIP - Recombinant human TNF alpha protein (ab9642)

Chromatin was prepared from HeLa (human epithelial cell line from cervix adenocarcinoma) cells treated with and without 20 ng/ml TNF- α (ab9642) for 60 minutes according to the Abcam X-ChIP protocol. Cells were fixed with formaldehyde for 10 minutes. The ChIP was performed with 25 μ g of chromatin, 5 μ g of [ab218533](#) (red), and 20 μ l of Protein A/G sepharose beads. 5 μ g of rabbit normal IgG was added to the beads control (gray). The immunoprecipitated DNA was quantified by real time PCR (SYBR green approach).

The ChIP data are consistent with the literature (PMID: 16135789).



Western blot - Recombinant human TNF alpha protein (ab9642)

All lanes : Anti-TNFAIP3 antibody [EPR2663] ([ab92324](#)) at 1/5000 dilution

Lane 1 : WEHI-3 (Mouse leukemia lymphoblast) whole cell lysate

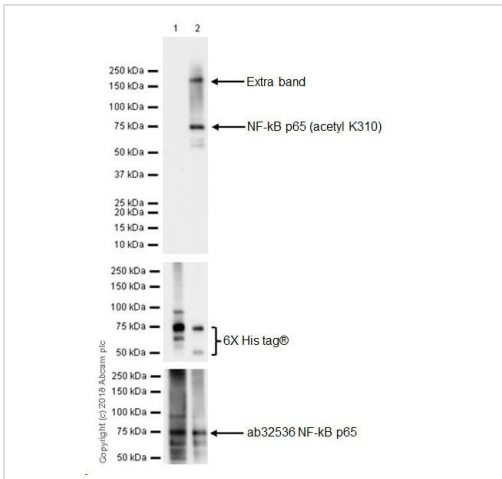
Lane 2 : WEHI-3 treated with 20 ng/ml TNF alpha (ab9642) for 6 h

Lysates/proteins at 20 μ g per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/20000 dilution

Observed band size: 80 kDa



Western blot - Recombinant human TNF alpha protein (ab9642)

All lanes : Anti-NF-kB p65 (acetyl K310) antibody [EPR21781] - ChIP Grade ([ab218533](#)) at 1/2000 dilution

Lane 1 : HEK-293 transfected with NF-kB p65 expression vector containing a myc-His-tag®, whole cell lysate

Lane 2 : HEK-293 transfected with NF-kB p65 and p300 (aa1287-1663) expression vectors containing a myc-His-tag®, then treated with 20 ng/ml TNF-alpha (ab9642) for 60 minutes, whole cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

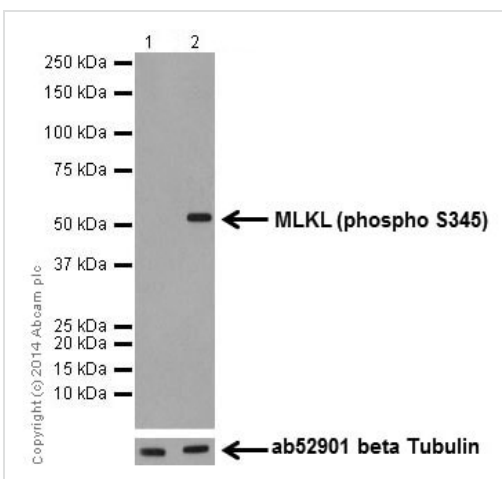
All lanes : Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/100000 dilution

Observed band size: 70 kDa

Exposure time: 37 seconds

Blocking/Dilution buffer: 5% NFDm/TBST.

NF-kB p65 (acetyl K310) expression is induced by TNF-α and p300 acetyltransferases (PMID: 20160011, PMID: 12456660, PMID: 16135789).



Western blot - Recombinant human TNF alpha protein (ab9642)

All lanes : Anti-MLKL (phospho S345) antibody [EPR9515(2)] ([ab196436](#)) at 1/1000 dilution

Lane 1 : Untreated L-929 (Mouse connective tissue fibroblast cells) whole cell lysate

Lane 2 : L-929 whole cell lysate treated with 20 ng/ml TNF alpha (ab9642), 100 nM Smac mimetic, and 20 µM z-VAD ([ab120382](#)) for 8 h and then harvested.

Lysates/proteins at 10 µg per lane.

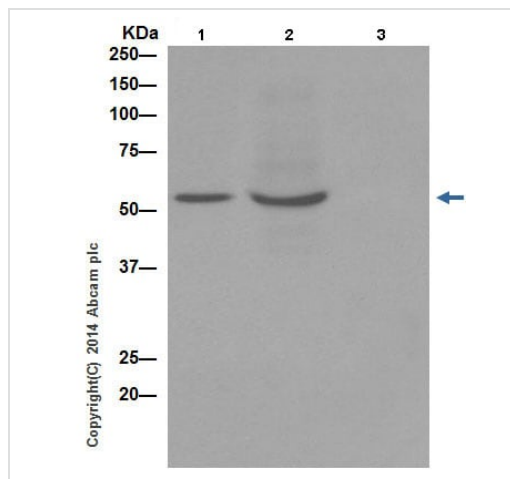
Secondary

All lanes : Peroxidase-conjugated goat anti-rabbit IgG (H+L) at 1/1000 dilution

Observed band size: 54 kDa

Exposure time: 15 seconds

Blocking and dilution buffer: 5% NFDm/TBST.



Immunoprecipitation - Recombinant human TNF alpha protein (ab9642)

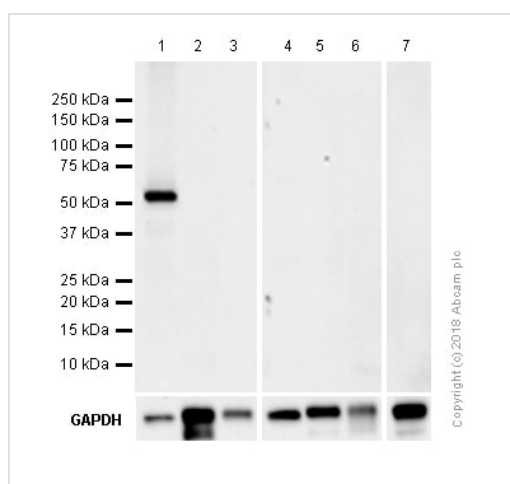
MLKL (phospho S345) was immunoprecipitated from 1 mg of L-929 (Mouse connective tissue fibroblast cells) whole cell lysate treated with 20 ng/ml TNF alpha (ab9642) + 100 nM Smac mimetic + 20 µM z-VAD compound (**ab120382**) for 8h using **ab196436** at 1/150 dilution. Western blot was performed from the immunoprecipitate using **ab196436** at 1/1000 dilution. Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG, was used as secondary antibody at 1/1500 dilution.

Lane 1: L-929 whole cell lysate treated with 20 ng/ml TNF alpha (ab9642) + 100 nM Smac mimetic+ 20 µM z-VAD compound (**ab120382**) for 8h; 10 µg (Input).

Lane 2: **ab196436** IP in L-929 whole cell lysate treated with 20 ng/ml TNF alpha (ab9642) + 100 nM Smac mimetic+ 20 µM z-VAD compound (**ab120382**) for 8h.

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of **ab196436** in L-929 whole cell lysate treated with 20 ng/ml TNF alpha (ab9642) + 100 nM Smac mimetic+ 20 µM z-VAD compound (**ab120382**) for 8h.

Blocking and dilution buffer and concentration: 5% NFDm/TBST.



Western blot - Recombinant human TNF alpha protein (ab9642)

All lanes : Anti-MLKL (phospho S345) antibody [EPR9515(2)] (**ab196436**) at 1/1000 dilution

Lane 1 : L-929 treated with 20 ng/ml TNF alpha (ab9642), 100 nM Smac mimetic, and 20 µM z-VAD (**ab120382**) for 8 h, whole cell lysate

Lane 2 : Mouse brain tissue lysate

Lane 3 : Mouse colon tissue lysate

Lane 4 : Mouse lung tissue lysate

Lane 5 : Mouse retina tissue lysate

Lane 6 : Mouse liver tissue lysate

Lane 7 : Raw264.7 (Mouse Abelson murine leukemia virus-induced tumor macrophage) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Observed band size: 54 kDa

Exposure time: 50 seconds

Blocking and diluting buffer: 5% NFDm/TBST.

MLKL pS345 is a trigger for necroptosis. It is only detectable in infection/cellular damaged (PMID:29229989) or aging tissue (PMID: 28807105) but not in normal tissues.

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