abcam

Product datasheet

Recombinant human IL-1 beta protein (Active) ab259387

★★★★★ 1 Abreviews 8 图像

描述

产品名称	重组人IL-1 beta蛋白(Active)
生物活性	Fully biologically active when compared to standard. ED_{50} is ≤ 0.7399 ng /ml, corresponding to a specific activity of 1.35 x 10^6 units/mg.
纯度	> 95 % SDS-PAGE. Purity by HPLC >=95%.
内毒素水平	<=0.005 Eu/µg
表达系统	HEK 293 cells
Accession	P01584
蛋白长度	Full length protein
无动物成分	Yes
无载体	是
性质	Recombinant
种属	Human
序列	APVRSLNCTL RDSQQKSLVM SGPYELKALH LQGQDMEQQV VFSMSFVQGE ESNDKIPVAL GLKEKNLYLS CVLKDDKPTL QLESVDPKNY PKKKMEKRFV FNKIEINNKL EFESAQFPNW YISTSQAENM PVFLGGTKGG QDITDFTMQF VSS
预测分子量	17 kDa
分子量信息	M + 1.02 Da (calc MW17433.98 Da)
氨基酸	117 to 269
额外的序列信息	N-Terminal Glycine. Full-length mature chain lacking the propeptide.

技术指标

Our **Abpromise guarantee** covers the use of **ab259387** in the following tested applications.

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

应用	Sandwich ELISA
	Cell Culture
	SDS-PAGE

Functional Studies

Mass Spectrometry

HPLC

形式 Lyophilized

补充说明 This protein is filter sterilised prior to aliquoting and lyophilisation. All aliquoting and lyophilisation steps are performed in a sterile environment

制备和贮存

稳定性和存储 Shipped at Room Temperature. Store at Room Temperature.

pH: 6.00
Constituents: 0.727% Dibasic monohydrogen potassium phosphate, 0.248% Monobasic dihydrogen potassium phosphate, 10.26% Trehalose

Buffer lyophilised from.

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

复溶 Reconstitute in PBS, aliquot and store at -80 C for 12 months or 4 C for 1 week. Avoid repeated freeze thaw. Lyophilized contents may appear as either a translucent film or a white powder. This variance does not affect the quality of the product.

常规信息

功能 Potent proinflammatory cytokine. Initially discovered as the major endogenous pyrogen, induces prostaglandin synthesis, neutrophil influx and activation, T-cell activation and cytokine production, B-cell activation and antibody production, and fibroblast proliferation and collagen production. Promotes Th17 differentiation of T-cells.

组织特异性 Expressed in activated monocytes/macrophages (at protein level).

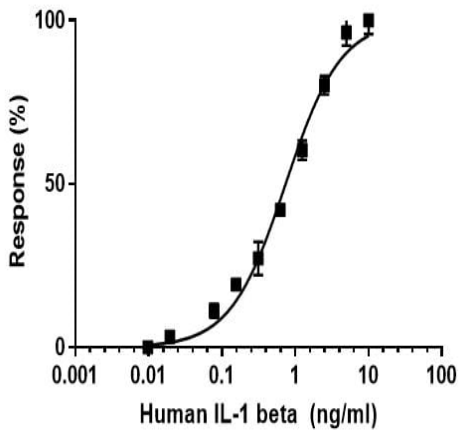
序列相似性 Belongs to the IL-1 family.

翻译后修饰 Activation of the IL1B precursor involves a CASP1-catalyzed proteolytic cleavage. Processing and secretion are temporarily associated.

细胞定位 Cytoplasm, cytosol. Lysosome. Secreted, exosome. Cytoplasmic vesicle, autophagosome. Secreted. The precursor is cytosolic. In response to inflammasome-activating signals, such as ATP for NLRP3 inflammasome or bacterial flagellin for NLRC4 inflammasome, cleaved and secreted. IL1B lacks any known signal sequence and the pathway(s) of its secretion is(are) not yet fully understood (PubMed:24201029). On the basis of experimental results, several unconventional secretion mechanisms have been proposed. 1. Secretion via secretory lysosomes: a fraction of CASP1 and IL1B precursor may be incorporated, by a yet undefined mechanism, into secretory lysosomes that undergo Ca(2+)-dependent exocytosis with release of mature IL1B (PubMed:15192144). 2. Secretory autophagy: IL1B-containing autophagosomes may fuse with endosomes or multivesicular bodies (MVBs) and then merge with the plasma membrane releasing soluble IL1B or IL1B-containing exosomes (PubMed:24201029). However, autophagy impacts IL1B production at several levels and its role in secretion is still controversial. 3. Secretion via exosomes: ATP-activation of P2RX7 leads to the formation of MVBs containing exosomes with entrapped IL1B, CASP1 and other inflammasome components. These MVBs undergo exocytosis with the release of exosomes. The release of soluble IL1B occurs after the lysis of exosome membranes (By similarity). 4. Secretion by microvesicle shedding: activation of

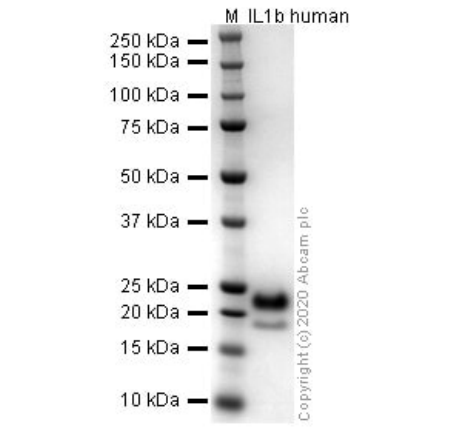
the ATP receptor P2RX7 may induce an immediate shedding of membrane-derived microvesicles containing IL1B and possibly inflammasome components. The cytokine is then released in the extracellular compartment after microvesicle lysis (PubMed:11728343). 5. Release by translocation through permeabilized plasma membrane. This may occur in cells undergoing pyroptosis due to sustained activation of the inflammasome (By similarity). These mechanisms may not be not mutually exclusive.

图片



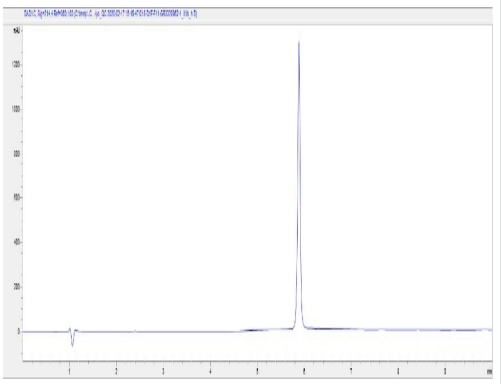
Functional Studies - Recombinant human IL-1 beta protein (Active) (ab259387)

Fully biologically active when compared to standard. ED_{50} is ≤ 0.7399 ng /ml, corresponding to a specific activity of 1.35 x 10^6 units/mg.



SDS-PAGE - Recombinant human IL-1 beta protein (Active) (ab259387)

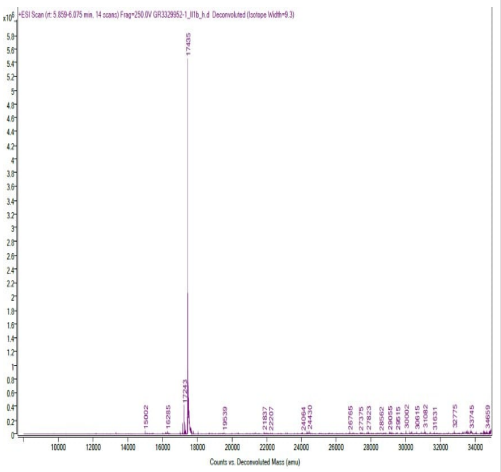
SDS-PAGE analysis of ab259387.



HPLC - Recombinant human IL-1 beta protein (Active) (ab259387)

Purity: 100%

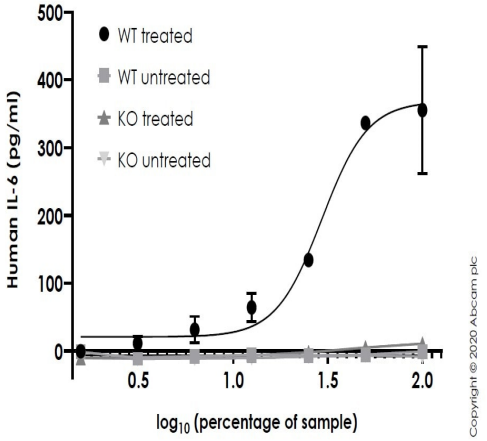
The spectrum was recorded using a 1260 Infinity II HPLC system with DAD and a MabPac RP column (3.0x100 mm, 4 µm). 5 µL of purified protein was injected and the gradient run from 80 % water:TFA (99.9:0.1 v/v) and 20 % acetonitrile:water:TFA (90:9.9:0.1 v/v/v) to 20 % water:TFA (99.9:0.1 v/v) and 80 % acetonitrile:water:TFA (90:9.9:0.1 v/v/v) within 3 minutes followed by an isocratic step for another 3 min. Flow rate was 0.5 mL/min and the column compartment temperature was 50 °C.



Mass Spectrometry - Recombinant human IL-1 beta protein (Active) (ab259387)

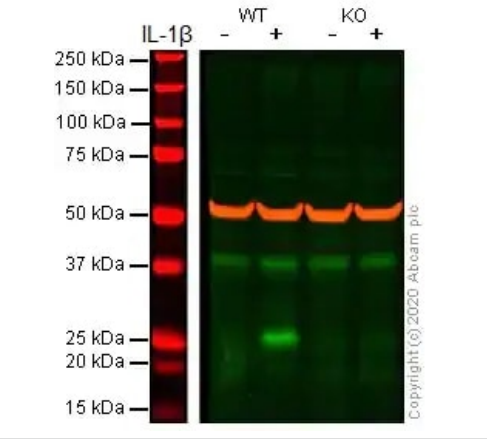
M + 1.02 Da (calc mass 17433.98)

The spectrum was recorded with a 6545XT AdvanceBio LC/Q-TOF (Agilent Technologies) and a MabPac RP column (42.1x50 mm, 4 µm, Thermo Scientific). 5 µL of purified protein was injected and the gradient run from 85 % water:FA (99.9:0.1 v/v) and 15 % acetonitrile:FA (90:9.9:0.1 v/v/v) to 55 % water:FA (99.9:0.1 v/v) and 45 % acetonitrile:FA (90:9.9:0.1 v/v/v) within 3 minutes followed by an isocratic step for another 2.5 min. Flow rate was 0.4 mL/min and the column compartment temperature was 60 °C. Data was analysed and deconvoluted using the Bioconfirm software (Agilent Technologies).



Sandwich ELISA - Recombinant human IL-1 beta protein (Active) (ab259387)

Human IL-6 concentration was interpolated from the IL-6 standard curve. Supernatants from cell culture samples were serially diluted and assessed by the Human IL-6 ELISA kit (**ab178013**). Wild-type and IL-6 knockout A549 cells (**ab273751**) were assessed in duplicate (n=2). Cells were either treated with 20 ng/mL active recombinant human IL-1 beta protein (ab259387) for 24 h to induce expression of IL-6 or not treated with IL-1 beta. Data are represented as the mean and error bars represent standard deviation.



Western blot - Recombinant human IL-1 beta protein (Active) (ab259387)

All lanes : Anti-IL-6 antibody [EPR21711] (**ab233706**) at 1/1000 dilution

Lane 1 : Wild-type A549 Brefeldin A (**ab120299**)-treated (5ug/ml, 4h) cell lysate
Lane 2 : Wild-type A549 IL-1ß (ab259387) (20 ng/ml, 24h) and Brefeldin A (**ab120299**)-treated (5 ug/ml for the last 4h) cell lysate
Lane 3 : IL-6 knockout A549 Brefeldin A (**ab120299**)-treated (5ug/ml, 4h) cell lysate
Lane 4 : IL-6 knockout A549 IL-1ß (ab259387) (20 ng/ml, 24h) and Brefeldin A (**ab120299**)-treated (5 ug/ml for the last 4h) cell lysate

Lysates/proteins at 30 µg per lane.

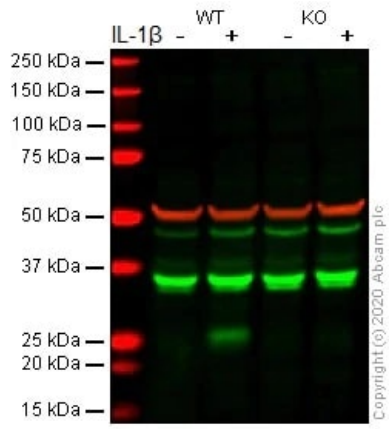
Performed under reducing conditions.

Observed band size: 25 kDa
Additional bands at: 40 kDa (possible non-specific binding)

Lanes 1 - 4: Merged signal (red and green). Green - **ab233706** observed at 25 kDa. Red - loading control **ab7291** (Mouse anti-Alpha Tubulin [DM1A] observed at 55kDa.

ab233706 was shown to react with IL-6 in wild-type A549 cells in western blot with loss of signal observed in IL-6 knockout cell line **ab273751** (knockout cell lysate **ab275501**). Wild-type and IL-6 knockout A549 cell lysates were subjected to SDS-PAGE. Membranes were blocked in fluorescent western blot (TBS-based)

blocking solution before incubation with **ab233706** and **ab7291** (Mouse anti-Alpha Tubulin [DM1A] overnight at 4°C at a 1 in 1000 Dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Recombinant human IL-1 beta protein (Active) (ab259387)

All lanes : Anti-IL-6 antibody [EPR20653] (**ab214429**) at 1/1000 dilution

Lane 1 : Wild-type A549 Brefeldin A (**ab120299**)-treated (5ug/ml, 4h) cell lysate
Lane 2 : Wild-type A549 IL-1ß (ab259387) (20 ng/ml, 24h) and Brefeldin A (**ab120299**)-treated (5 ug/ml for the last 4h) cell lysate
Lane 3 : IL-6 knockout A549 Brefeldin A (**ab120299**)-treated (5ug/ml, 4h) cell lysate
Lane 4 : IL-6 knockout A549 IL-1ß (ab259387) (20 ng/ml, 24h) and Brefeldin A (**ab120299**)-treated (5 ug/ml for the last 4h) cell lysate

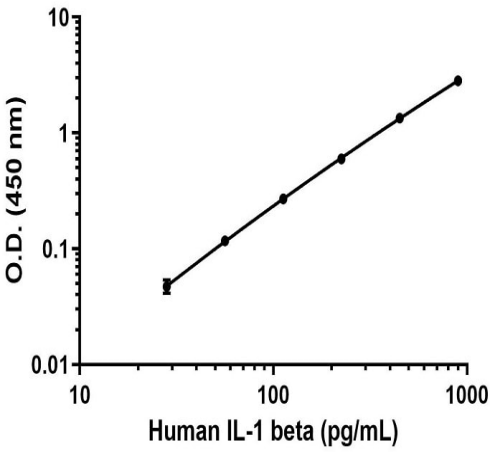
Lysates/proteins at 30 µg per lane.

Performed under reducing conditions.

Observed band size: 25 kDa
Additional bands at: 35 kDa (possible non-specific binding)

Lanes 1 - 4: Merged signal (red and green). Green - **ab214429** observed at 25 kDa. Red - loading control **ab7291** (Mouse anti-Alpha Tubulin [DM1A] observed at 55kDa.

ab214429 was shown to react with IL-6 in wild-type A549 cells in western blot with loss of signal observed in IL-6 knockout cell line **ab273751** (knockout cell lysate **ab275501**). Wild-type A549 and IL-6 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with **ab214429** and **ab7291** (Mouse anti-Alpha Tubulin [DM1A] overnight at 4°C at a 1 in 1000 Dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Sandwich ELISA - Recombinant human IL-1 beta protein (Active) (ab259387)

Background subtracted standard curve using Human IL-1 beta Antibody Pair - BSA and Azide free (**ab241807**) and Recombinant human IL-1 beta protein (Active) (ab259387) in sandwich ELISA. The ELISA was performed using the components of the corresponding SimpleStep® kit, which uses the same antibody pair with a different formulation and format.

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