# abcam

## Product datasheet

## Anti-TGF beta 1 antibody [EPR21143] ab215715





★★★★★ 6 Abreviews 134 References 10 Images

#### Overview

**Product name** Anti-TGF beta 1 antibody [EPR21143]

**Description** Rabbit monoclonal [EPR21143] to TGF beta 1

**Host species** Rabbit

Specificity For testing samples with low expression level of TGF beta 1, we recommend ab179695 which

could give stronger signal. Loading larger amount of lysate or lower antibody dilution would also

help.

**Tested applications** Suitable for: WB, IHC-P

Species reactivity Reacts with: Mouse, Rat, Human

**Immunogen** Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: NIH/3T3, L-929, HeLa, A549, HL-60, RAW 264.7, Wild-type A549, K562 and SH-SY5Y

whole cell lysates; Mouse and Rat spleen lysate; Mouse heart tissue lysate; C6 cell lysate. IHC-P:

Human thrombocytosis tissue; human bone; Mouse and rat spleen tissues.

**General notes** This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility

- Improved sensitivity and specificity

- Long-term security of supply

- Animal-free production

For more information see here.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**® **patents**.

#### **Properties**

**Form** Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long

term. Avoid freeze / thaw cycle.

Storage buffer pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: PBS, 40% Glycerol, 0.05% BSA

**Purity** Protein A purified

ClonalityMonoclonalClone numberEPR21143

**Isotype** IgG

## **Applications**

The Abpromise guarantee

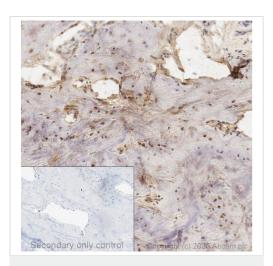
Our <u>Abpromise guarantee</u> covers the use of ab215715 in the following tested applications.

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

Application	Abreviews	Notes
WB	****(1)	1/1000. Predicted molecular weight: 44 kDa. Actual band: 13 kDa and 44 kDa
IHC-P	<b>★★★★★</b> (4)	1/500. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

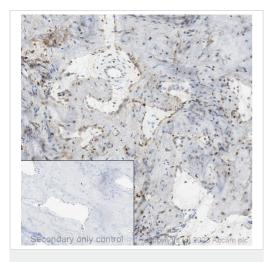
Target		
Function	Multifunctional protein that controls proliferation, differentiation and other functions in many cell types. Many cells synthesize TGFB1 and have specific receptors for it. It positively and negatively regulates many other growth factors. It plays an important role in bone remodeling as it is a potent stimulator of osteoblastic bone formation, causing chemotaxis, proliferation and differentiation in committed osteoblasts.	
Tissue specificity	Highly expressed in bone. Abundantly expressed in articular cartilage and chondrocytes and is increased in osteoarthritis (OA). Co-localizes with ASPN in chondrocytes within OA lesions of articular cartilage.	
Involvement in disease	Defects in TGFB1 are the cause of Camurati-Engelmann disease (CE) [MIM:131300]; also known as progressive diaphyseal dysplasia 1 (DPD1). CE is an autosomal dominant disorder characterized by hyperostosis and sclerosis of the diaphyses of long bones. The disease typically presents in early childhood with pain, muscular weakness and waddling gait, and in some cases other features such as exophthalmos, facial paralysis, hearing difficulties and loss of vision.	
Sequence similarities	Belongs to the TGF-beta family.	
Post-translational modifications	Glycosylated.  The precursor is cleaved into mature TGF-beta-1 and LAP, which remains non-covalently linked to mature TGF-beta-1 rendering it inactive.	
Cellular localization	Secreted > extracellular space > extracellular matrix.	

## **Images**



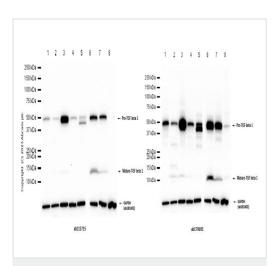
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TGF beta 1 antibody
[EPR21143] (ab215715)

Immunohistochemical analysis of formalin-fixed paraffin-embedded human bone labelling TGF beta 1 with ab215715 at a concentration of 3µg/ml. The immunostaining was performed on a Leica Biosystems BOND<sup>®</sup> RX instrument with a Bond ™ Polymer Refine Detection kit. Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution 2) for 20 mins. ab215715 anti-TGF beta 1 antibody [EPR21143] was incubated for 15mins at room temperature. Sections were counterstained with Hematoxylin. Image inset shows absence of staining in secondary antibody only control.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TGF beta 1 antibody
[EPR21143] (ab215715)

Immunohistochemical analysis of formalin-fixed paraffin-embedded human bone labelling TGF beta 1 with ab215715 at a dilution of 4µg/ml. The immunostaining was performed on a Ventana DISCOVERY ULTRA (Roche Tissue Diagnostics) instrument with an OptiView DAB IHC Detection Kit. Heat mediated antigen retrieval was conducted for 32min with ULTRA cell conditioning solution (CC1 pH8.5). ab215715 anti TGF beta 1 antibody was incubated at 37°C for 16min. Sections were counterstained is with Hematoxylin II. Image inset shows absence of staining in secondary antibody only control.



Western blot - Anti-TGF beta 1 antibody [EPR21143] (ab215715)

**All lanes :** Anti-TGF beta 1 antibody [EPR21143] (ab215715) at 1/1000 dilution

**Lane 1 :** A549 (Human lung carcinoma epithelial cell) whole cell lysate at 20  $\mu$ g

Lane 2 : HL-60 (Human acute promyelocytic leukemia promyeloblast) whole cell lysate at 20 µg

**Lane 3**: K-562 (Human chronic myelogenous leukemia lymphoblast) whole cell lysate at 20 μg

Lane 4: NIH/3T3 (Mouse embryonic fibroblast) whole cell lysate at 20 µg

Lane 5: C6 (Rat glial tumor glial cell) whole cell lysate at 20  $\mu g$ 

Lane 6 : Mouse spleen tissue lysateLane 7 : Rat spleen tissue lysateLane 8 : Mouse heart tissue lysate

## Secondary

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

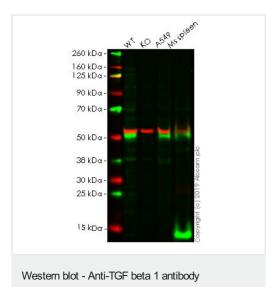
Predicted band size: 44 kDa Observed band size: 12.44 kDa

Exposure time: 20 seconds

Blocking and diluting buffer and concentration: 5% NFDM/TBST.

ab181602 was used as a GAPDH loading control.

For testing samples with low expression level of TGF beta 1, we recommend <u>ab179695</u> which could give stronger signal. Loading larger amount of lysate or lower antibody dilution would also help.



[EPR21143] (ab215715)

**All lanes :** Anti-TGF beta 1 antibody [EPR21143] (ab215715) at 1/1000 dilution

Lane 1: Wild-type HeLa whole cell lysate

Lane 2: TGFB1 knockout HeLa whole cell lysate

Lane 3: A549 whole cell lysate

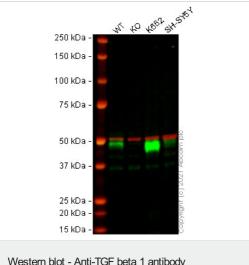
Lane 4: Mouse spleen tissue lysate

Lysates/proteins at 20 µg per lane.

**Predicted band size:** 44 kDa **Observed band size:** 13,44 kDa

**Lanes 1 - 4:** Merged signal (red and green). Green - ab215715 observed at 13 and 44 kDa. Red - loading control, <u>ab7291</u>, observed at 50 kDa.

ab215715 was shown to specifically react with in wild-type HeLa cells as signal was lost in TGFB1 knockout cells. Wild-type and TGFB1 knockout samples were subjected to SDS-PAGE. The membrane was blocked with 3% NF Milk. Ab215715 and ab7291 (Mouse anti-tubulin loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/20000 dilution respectively. Blots were developed 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 1/20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-TGF beta 1 antibody [EPR21143] (ab215715)

**All lanes :** Anti-TGF beta 1 antibody [EPR21143] (ab215715) at 1/1000 dilution

Lane 1: Wild-type A549 cell lysate

Lane 2: TGFB1 knockout A549 cell lysate

Lane 3 : K562 cell lysate

Lane 4 : SH-SY5Y cell lysate

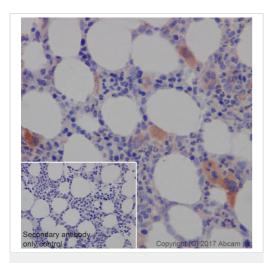
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 44 kDa Observed band size: 48 kDa

**Lanes 1 - 4:** Merged signal (red and green). Green - ab215715 observed at 48 kDa. Red - loading control <u>ab7291</u> (Mouse anti-Alpha Tubulin [DM1A]) observed at 55 kDa.

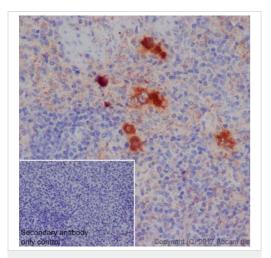
ab215715 was shown to react with TGF beta 1 in wild-type A549 cells in Western blot with loss of signal observed in TGFB1 knockout cell line <a href="mailto:ab269509">ab269509</a> (TGFB1 knockout cell lysate <a href="mailto:ab269671">ab269671</a>). Wild-type A549 and TGFB1 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with ab215715 and <a href="mailto:ab7291">ab7291</a> (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 lgG H&L (IRDye® 800CW) preabsorbed (<a href="mailto:ab216773">ab216773</a>) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preabsorbed (<a href="mailto:ab216776">ab216773</a>) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TGF beta 1 antibody
[EPR21143] (ab215715)

Immunohistochemical analysis of paraffin-embedded human thrombocytosis tissue labeling TGF beta 1 with ab215715 at 1/500 dilution, followed by Goat Anti-Rabbit lgG H&L (HRP) ready to use. Cytoplasmic staining of megakaryocytes in human thrombocytosis (PMID: 25305163). Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

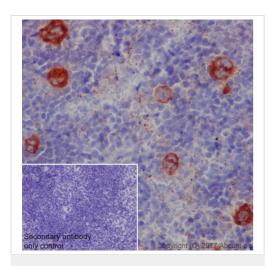
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) ready to use.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TGF beta 1 antibody
[EPR21143] (ab215715)

Immunohistochemical analysis of paraffin-embedded rat spleen tissue labeling TGF beta 1 with ab215715 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) ready to use. Positive staining of megakaryocytes and platelets in rat spleen (PMID: 25305163). Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

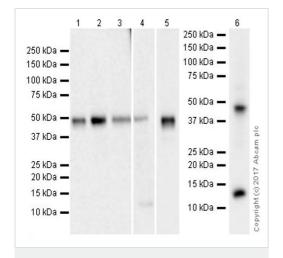
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) ready to use.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TGF beta 1 antibody
[EPR21143] (ab215715)

Immunohistochemical analysis of paraffin-embedded mouse spleen tissue labeling TGF beta 1 with ab215715 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) ready to use. Positive staining of megakaryocytes and platelets in mouse spleen (PMID: 25305163). Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) ready to use.



Western blot - Anti-TGF beta 1 antibody [EPR21143] (ab215715)

**All lanes :** Anti-TGF beta 1 antibody [EPR21143] (ab215715) at 1/1000 dilution

Lane 1 : NIH/3T3 (mouse embryo fibroblast cell line) whole cell lysate at 20 µg

**Lane 2**: L-929 (mouse connective tissue fibroblast cell line) whole cell lysate at 20  $\mu g$ 

**Lane 3 :** A549 (human lung carcinoma cell line) whole cell lysate at 20  $\mu g$ 

**Lane 4 :** HL-60 (human promyelocytic leukemia cell line) whole cell lysate at 20  $\mu g$ 

**Lane 5 :** RAW 264.7 (mouse macrophage cell line transformed with Abelson murine leukemia virus) whole cell lysate at 20  $\mu$ g

Lane 6: Mouse spleen lysate at 10 µg

#### Secondary

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

Developed using the ECL technique.

**Predicted band size:** 44 kDa **Observed band size:** 12,44 kDa

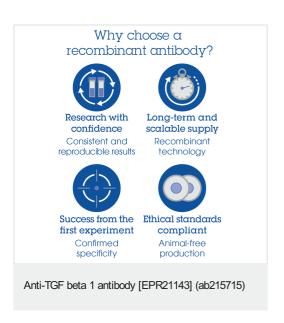
Exposure times: Lanes 1-4: 3 minutes; Lane 5: 41 seconds; Lane

6:24 seconds.

Blocking/Dilution buffer: 5% NFDM/TBST.

The molecular mass observed is consistent with the literature (PMID 2139036).

The blot was developed on a BIO-RAD® ChemiDoc™ MP instrument.



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