

Product datasheet

Anti-TCF-4 antibody [NCI-R159-6] ab217668

Recombinant RabMAb

★★★★★ [1 Abreviews](#) [25 References](#) [9 Images](#)

Overview

Product name	Anti-TCF-4 antibody [NCI-R159-6]
Description	Rabbit monoclonal [NCI-R159-6] to TCF-4
Host species	Rabbit
Specificity	Mouse specificity in WB only. Weak human specificity in IHC.
Tested applications	Suitable for: Flow Cyt (Intra), WB, ChIP, IHC
Species reactivity	Reacts with: Mouse, Human
Immunogen	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: Nuclear extracts of Cal-1 cells infected with control TCF-4 shRNA. SH-SY5Y nuclear extracts. U-87 MG and Neuro-2a whole cell lysates, Daudi nuclear fraction, U-87 MG whole cell lysate, Neuro-2a whole cell lysate IHC: Human tonsil. Blastic plasmacytoid dendritic cell neoplasm (BPDCN) cell pellets after selection and induction of shRNA expression for 1 day. Flow Cyt (intra): Cal-1 cells. ChIP: Cal-1 cells.
General notes	<p>For detailed protocol using this antibody for IHC, ChIP, and Flow Cyt, please refer to the following paper:</p> <p><u>A Druggable TCF4- and BRD4-Dependent Transcriptional Network Sustains Malignancy in Blastic Plasmacytoid Dendritic Cell Neoplasm</u></p> <p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none">- High batch-to-batch consistency and reproducibility- Improved sensitivity and specificity- Long-term security of supply- Animal-free production <p>For more information see here.</p> <p>Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb[®] patents.</p>

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: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

Purity

Protein A purified

Clonality

Monoclonal

Clone number

NCI-R159-6

Isotype

IgG

Applications**The Abpromise guarantee**

Our **Abpromise guarantee** covers the use of ab217668 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
Flow Cyt (Intra)		1/100.
WB		1/10000. Detects a band of approximately 90 kDa (predicted molecular weight: 71 kDa).
ChIP		Use a concentration of 2.5 µg/ml.
IHC		1/100. Fixative: 4% Formalin Antigen retrieval: Low pH retrieval solution

Target**Function**

Transcription factor that binds to the immunoglobulin enhancer Mu-E5/KE5-motif. Binds to the E-box present in the somatostatin receptor 2 initiator element (SSTR2-ISR) to activate transcription (By similarity). Preferentially binds to either 5'-ACANNTGT-3' or 5'-CCANNTGG-3'.

Tissue specificity

Expressed in adult heart, brain, placenta, skeletal muscle and to a lesser extent in the lung. In developing embryonic tissues, expression mostly occurs in the brain.

Involvement in disease

Defects in TCF4 are a cause of Pitt-Hopkins syndrome (PTHS) [MIM:610954]. PTHS is a rare syndromic encephalopathy characterized by severe psychomotor delay, epilepsy, daily bouts of diurnal hyperventilation starting in infancy, mild postnatal growth retardation, postnatal microcephaly, and distinctive facial features. Since most hitherto reported cases have been sporadic, with males and females equally affected, PTHS is regarded as an autosomal dominant condition.

Sequence similarities

Contains 1 basic helix-loop-helix (bHLH) domain.

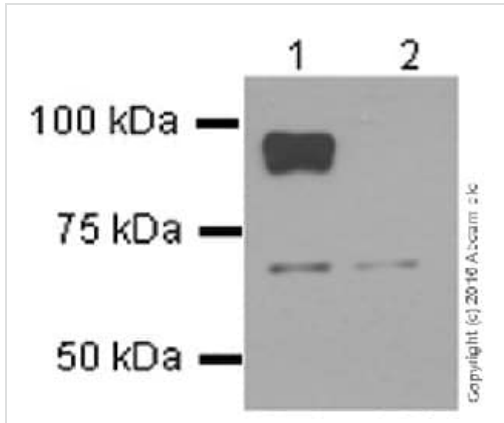
Domain

the 9aaTAD motif is a transactivation domain present in a large number of yeast and animal transcription factors.

Cellular localization

Nucleus.

Images



Western blot - Anti-TCF-4 antibody [NCI-R159-6] (ab217668)

All lanes : Anti-TCF-4 antibody [NCI-R159-6] (ab217668) at 1/10000 dilution

Lane 1 : Nuclear extracts of Cal-1 cells (Human plasmacytoid dendritic cell line) infected with control TCF4 shRNA

Lane 2 : Nuclear extracts of Cal-1 cells infected with TCF4 shRNA

Lysates/proteins at 20 µg per lane.

Secondary

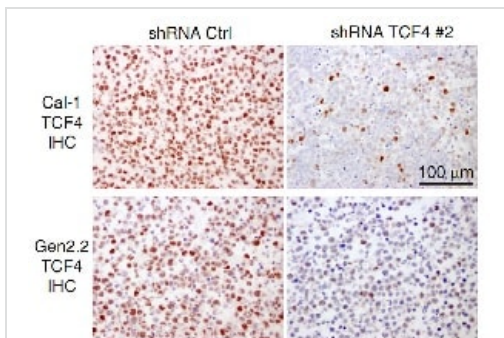
All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated

Predicted band size: 71 kDa

Observed band size: 90 kDa

Exposure time: 40 seconds

The data was provided by the collaborator Dr. Louis M. Staudt, NCI, NIH.

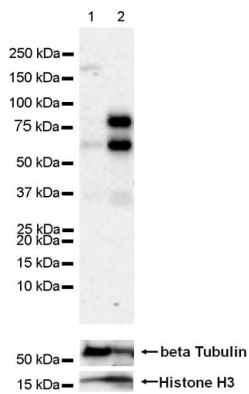


Immunohistochemistry - Anti-TCF-4 antibody [NCI-R159-6] (ab217668)

Immunohistochemical analysis of 4% Formalin fixed Blastic plasmacytoid dendritic cell neoplasm (BPDCN) cell pellets after selection and induction of shRNA expression for 1 day, labeling TCF-4 with ab217668 at 1/100 dilution. Universal DAB Detection Kit was used for detection of IHC staining on an automated system.

The data was provided by our collaborator Dr. Louis M. Staudt, and published in Cancer Cell 30, 764-778, 2016 (PMID: 27846392).

Several TCF-4 shRNAs were used. This IHC image shows data for shRNA TCF4 #2.



Western blot - Anti-TCF-4 antibody [NCI-R159-6] (ab217668)

All lanes : Anti-TCF-4 antibody [NCI-R159-6] (ab217668) at 1/1000 dilution

Lane 1 : Daudi (human Burkitt's lymphoma lymphoblast) cytoplasmic fraction

Lane 2 : Daudi nuclear fraction

Lysates/proteins at 20 µg per lane.

Secondary

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

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 71 kDa

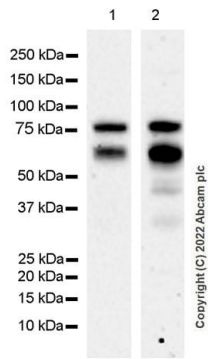
Observed band size: 58,79 kDa

Exposure time: 3 minutes

Exposure time : 3 minutes

Blocking/Dilution buffer: 5% NFDm/TBST.

This blot was developed using a high sensitivity ECL substrate.



Western blot - Anti-TCF-4 antibody [NCI-R159-6] (ab217668)

All lanes : Anti-TCF-4 antibody [NCI-R159-6] (ab217668) at 1/1000 dilution

Lane 1 : U-87 MG (human glioblastoma-astrocytoma epithelial cell), whole cell lysate

Lane 2 : Neuro-2a (mouse neuroblastoma neuroblast), whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

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

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 71 kDa

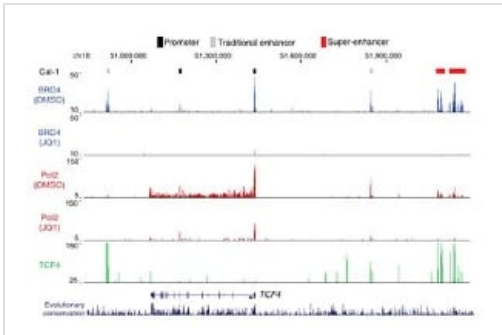
Observed band size: 58,79 kDa

Exposure time: 3 minutes

Exposure time : 3 minutes

Blocking/Dilution buffer: 5% NFDm/TBST.

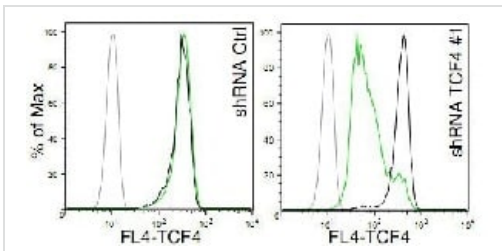
This blot was developed using a high sensitivity ECL substrate.



ChIP - Anti-TCF-4 antibody [NCI-R159-6] (ab217668)

Cal-1 cells (Human plasmacytoid dendritic cell line) were cross-linked with 1% formaldehyde for 5 min at RT. Cross-linked cells were first washed with ice-cold PBS and then resuspended in ice-cold RIPA buffer (10mM Tris-HCl pH8, 140 mM NaCl, 1mM EDTA pH 8, 0.5 mM EGTA, 1% Triton X-100, 0.1% SDS and 0.1% Sodium Deoxycholate) to a final concentration of 5×10^6 cells/ml. DNA was sheared with a Misonix XL sonicator, by performing 12 x 45" sonication cycles at power setting of 5. For each ChIP reaction, 2×10^7 chromatin cell equivalents were incubated overnight with $10 \mu\text{g}$ of ab217668. The following day, chromatin/antibody complexes were incubated with $50 \mu\text{l}$ of Protein G/Protein A magnetic beads mix (G to A ratio 3:1) for 4 h at 4°C . Normal rabbit IgG was added to the beads as control.

The TCF-4 locus ChIP-seq tracks for BRD4 (blue), RNA Pol2 (red), and TCF-4 (green) are shown for Cal-1 cells. This data was kindly provided by our collaborator Dr. Louis M. Staudt, and has been published (PMID: 27846392).

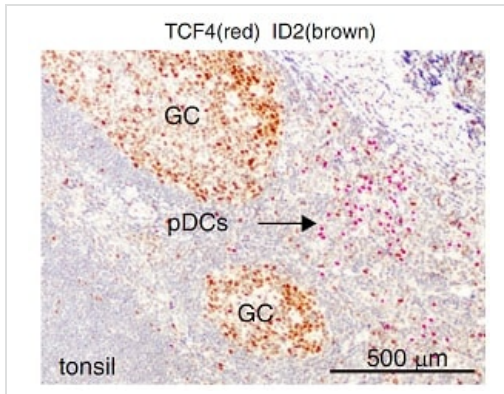


Flow Cytometry (Intracellular) - Anti-TCF-4 antibody [NCI-R159-6] (ab217668)

Intracellular flow cytometric analysis of 1% paraformaldehyde-fixed, ice-cold methanol-permeabilized Cal-1 cells (Human plasmacytoid dendritic cell line) (black - positive control) and Cal-1 cells infected with either Ctrl (left green) or TCF-4 (right green) shRNA, labeling TCF-4 with ab217668 at 1/100 dilution (green and black) compared with a Rabbit IgG control (grey). Goat anti-Rabbit IgG (Alexa Fluor[®] 647) at 1/500 dilution was used as the secondary antibody.

The data was provided by our collaborator Dr. Louis M. Staudt, and published in Cancer Cell 30, 764-778, 2016 (PMID: 27846392). Several TCF4 shRNAs were used. This FC image shows data for shRNA TCF4

1.



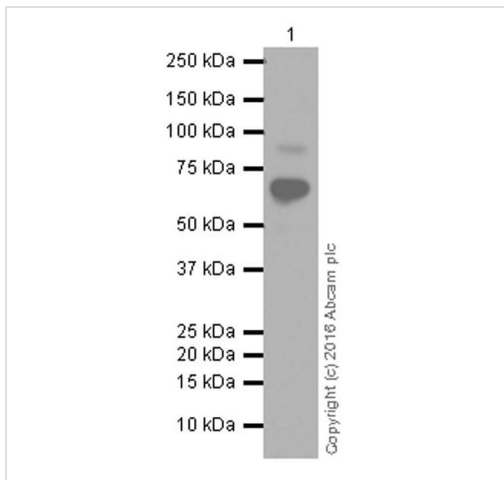
Immunohistochemistry - Anti-TCF-4 antibody [NCI-R159-6] (ab217668)

Immunohistochemical analysis of 4% Formalin fixed human tonsil labeling TCF-4 with ab217668 at 1/100 dilution. Universal DAB Detection Kit was used for detection of IHC staining on an automated system.

pDCs: plasmacytoid dendritic cells.

GC: Germinal Center.

The data was provided by our collaborator Dr. Louis M. Staudt, and published in Cancer Cell 30, 764-778, 2016 (PMID: 27846392).



Western blot - Anti-TCF-4 antibody [NCI-R159-6] (ab217668)

Anti-TCF-4 antibody [NCI-R159-6] (ab217668) at 1/200 dilution + SH-SY5Y (Human neuroblastoma cell line from bone marrow) nuclear extracts at 10 μg

Secondary

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

Predicted band size: 71 kDa

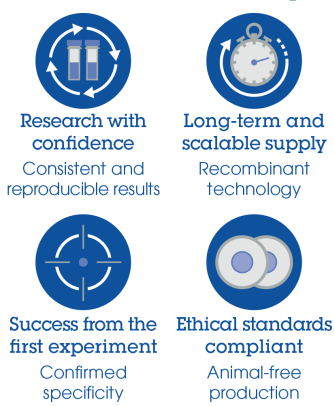
Observed band size: 90 kDa

Exposure time: 3 minutes

Blocking and Diluting buffer and concentration: 5% NFDM /TBST

The isoforms expression pattern is consistent with the literatures (PMID: 21789225).

Why choose a recombinant antibody?



Research with confidence
Consistent and reproducible results

Long-term and scalable supply
Recombinant technology

Success from the first experiment
Confirmed specificity

Ethical standards compliant
Animal-free production

Anti-TCF-4 antibody [NCI-R159-6] (ab217668)

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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