abcam

ab223073

Anti-TCF-4 antibody [NCI-R159-6] - BSA and Azide free

Recombinant | RabMAb | Advanced Validation

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	NCI-R159-6
Purification technique	Affinity purification Protein A
Specificity	Mouse specificity in WB only.Weak human specificity in IHC.
Concentration	1.05 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

ChIC/CUT&RUN-seq

Tested

Species	Mouse, Human

Dilution info	-
Notes	-

ChIP

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Mouse
Dilution info	-
Notes	-

IHC-P

Tested

Species	Human
Dilution info	-
Notes	Fixative: 4% Formalin Antigen retrieval: Low pH retrieval solution.

Not recommended

Species	Mouse
Dilution info	-
Notes	Fixative: 4% Formalin Antigen retrieval: Low pH retrieval solution

WB

Tested

Species	Human, Mouse

Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Mouse
Dilution info	-
Notes	-

Target data

See full target information TCF4

Function	Transcription factor that binds to the immunoglobulin enhancer Mu-E5/KE5-motif. Involved in the initiation of neuronal differentiation. Activates transcription by binding to the E box (5'-CANNTG-3'). Binds to the E-box present in the somatostatin receptor 2 initiator element (SSTR2-INR) to activate transcription (By similarity). Preferentially binds to either 5'-ACANNTGT-3' or 5'-CCANNTGG-3'.

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C
Storage information	Do Not Freeze

Notes

ab223073 is the carrier-free version of ab217668.

For detailed protocol using this antibody for IHC, ChIP, and Flow Cyt, please refer to the following paper: Ceribelli et al. A Druggable TCF4- and BRD4-Dependent Transcriptional Network Sustains Malignancy in Blastic Plasmacytoid Dendritic Cell Neoplasm, PMID: 27846392

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

What are the advantages of a recombinant monoclonal antibody?
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 batch production

For more information, read more on recombinant antibodies.

Conjugation ready
Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with 1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

Compatibility
This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

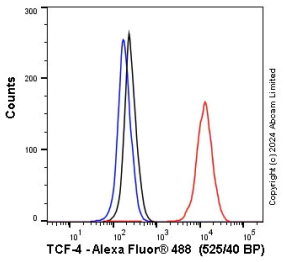
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

15 product images



Flow Cytometry (Intracellular) - Anti-TCF-4 antibody [NCI-R159-6] - BSA and Azide free (ab223073)

TCF-4 Flow Cytometry (Intracellular) staining of SH-SY5Y cells using rabbit Anti-TCF-4 antibody

This data was developed using ab217668, the same antibody clone in a different buffer formulation.

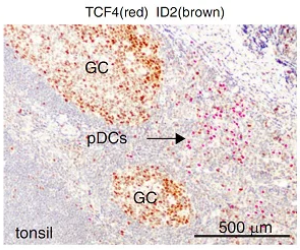
Flow cytometry overlay histogram showing SH-SY5Y cells stained with ab217668 (red line). The cells were fixed with 4% formaldehyde (10 min) and then permeabilised with 0.1% PBS-Triton X-100 for 15 min. The cells were incubated in nan followed by the antibody (ab217668) (1x 10^6in 100µl at 0.1µg/ml (1/19400)) for 30min at 22°C.

The secondary antibody Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed was incubated at 1/4000 for 30min at 22°C

Isotype control antibody (black line) was Recombinant Rabbit IgG, monoclonal [EPR25A] - Isotype Control used at the same concentration and conditions as the primary antibody. Unlabelled sample (blue line) was also used as a control.

Acquisition of >5000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter.

This antibody gave a positive signal in SH-SY5Y Fixed with 80% methanol (5 min) / permeabilised with 0.1% PBS-Triton X-100 for 15 min under the same conditions.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-TCF-4 antibody [NCI-R159-6] - BSA and Azide free (ab223073)

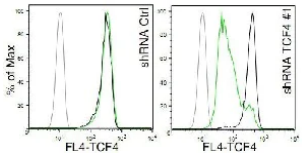
TCF-4 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-TCF-4 antibody

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).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab217668).



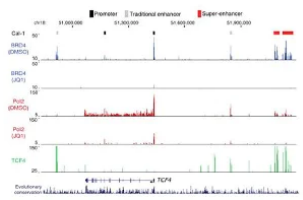
Flow Cytometry (Intracellular) - Anti-TCF-4 antibody [NCI-R159-6] - BSA and Azide free (ab223073)

TCF-4 Flow Cytometry (Intracellular) staining using rabbit Anti-TCF-4 antibody

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 TCF4 (right green) shRNA, labeling TCF4 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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab217668).

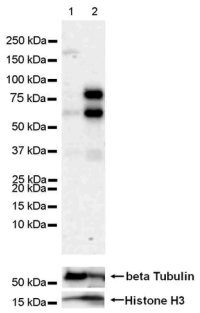


ChIP - Anti-TCF-4 antibody [NCI-R159-6] - BSA and Azide free (ab223073)

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 $5x10^6$ 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, $2x10^7$ chromatin cell equivalents were incubated overnight with10 µg of ab217668. The following day, chromatin/antibody complexes were incubated with 50 µl 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).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab217668).



Western blot - Anti-TCF-4 antibody [NCI-R159-6] - BSA and Azide free (ab223073)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab217668).
Blocking/Dilution buffer: 5% NFDM/TBST.

This blot was developed using a high sensitivity ECL substrate. The high-sensitivity ECL substrate used allows for the detection of proteins in the mid-femtogram range.

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

Lane 1:
Daudi (human Burkitt's lymphoma lymphoblast) cytoplasmic fraction at 20 µg

Lane 2:
Daudi nuclear fraction at 20 µg

Secondary

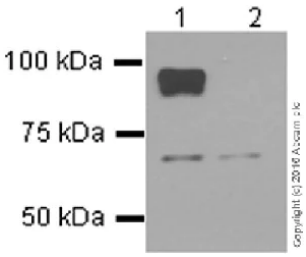
All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Developed using the ECL technique.

Predicted band size: 71 kDa

Observed band size: 58 kDa, 79 kDa

Exposure time: 3min



Western blot - Anti-TCF-4 antibody [NCI-R159-6] - BSA and Azide free (ab223073)

TCF-4 Western blot staining using rabbit Anti-TCF-4 antibody

This data was developed using ab217668, the same antibody clone in a different buffer formulation.
The data was provided by the collaborator Dr. Louis M. Staudt, NCI, NIH.

All lanes:
Western blot - 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 at 20 µg

Lane 2:
Nuclear extracts of Cal-1 cells infected with TCF4 shRNA at 20 µg

Secondary

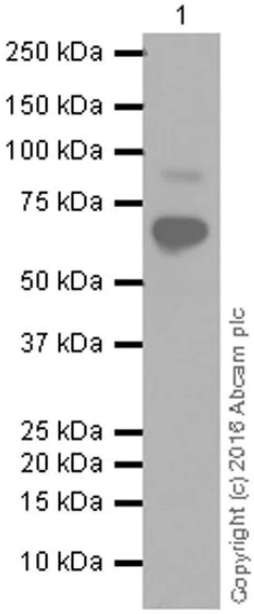
All lanes:

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

Predicted band size: 71 kDa

Observed band size: 90 kDa

Exposure time: 40s



Western blot - Anti-TCF-4 antibody [NCI-R159-6] - BSA and Azide free (ab223073)

TCF-4 Western blot staining of SH-SY5Y (Human neuroblastoma cell line from bone marrow) nuclear extracts using rabbit Anti-TCF-4 antibody

This data was developed using ab217668, the same antibody clone in a different buffer formulation.
Blocking and Diluting buffer and concentration: 5% NFDM /TBST.

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

All lanes:
Western blot - Anti-TCF-4 antibody [NCI-R159-6] (ab217668) at 1/200 dilution

All lanes:
SH-SY5Y (Human neuroblastoma cell line from bone marrow) nuclear extracts at 10 µg

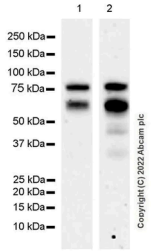
Secondary

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

Predicted band size: 71 kDa

Observed band size: 90 kDa

Exposure time: 3min



Western blot - Anti-TCF-4 antibody [NCI-R159-6] - BSA and Azide free (ab223073)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab217668).
Blocking/Dilution buffer: 5% NFDM/TBST.

This blot was developed using a high sensitivity ECL substrate. The high-sensitivity ECL substrate used allows for the detection of proteins in the mid-femtogram range.

All lanes:
Western blot - 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 at 20 µg

Lane 2:
Neuro-2a (mouse neuroblastoma neuroblast), whole cell lysate at 20 µg

Secondary

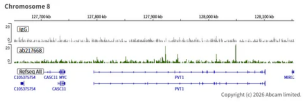
All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Developed using the ECL technique.

Predicted band size: 71 kDa

Observed band size: 58 kDa, 79 kDa

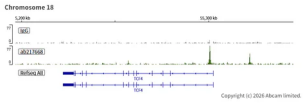
Exposure time: 3min



ChIC/CUT&RUN sequencing - Anti-TCF-4 antibody [NCI-R159-6] - BSA and Azide free (ab223073)

This data was developed using ab217668, the same antibody clone in a different buffer formulation.

ChIC/CUT&RUN was performed using a pAG-MNase at a final concentration of 700 ng/mL, 2.5 x 10^5 SK-N-SH (human neuroblastoma epithelial cell) cells and 5 µg of ab217668 [NCI-R159-6]. The resulting DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 10 million reads. The negative IgG control ab172730 is also shown. The University of Geneva owns patents relevant to ChIC (Chromatin Immuno-Cleavage) methods.



ChIC/CUT&RUN sequencing - Anti-TCF-4 antibody [NCI-R159-6] - BSA and Azide free (ab223073)

This data was developed using ab217668, the same antibody clone in a different buffer formulation.

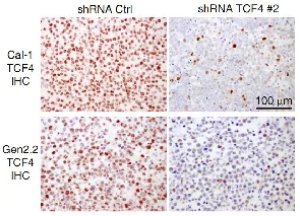
ChIC/CUT&RUN was performed using a pAG-MNase at a final concentration of 700 ng/mL, 2.5 x 10^5 SK-N-SH (human neuroblastoma epithelial cell) cells and 5 µg of ab217668 [NCI-R159-6]. The resulting DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 10 million reads. The negative IgG control ab172730 is also shown. The University of Geneva owns patents relevant to ChIC (Chromatin Immuno-Cleavage) methods.



ChIC/CUT&RUN sequencing - Anti-TCF-4 antibody [NCI-R159-6] - BSA and Azide free (ab223073)

This data was developed using ab217668, the same antibody clone in a different buffer formulation.

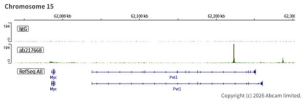
ChIC/CUT&RUN was performed using a pAG-MNase at a final concentration of 700 ng/mL, 2.5 x 10^5 SK-N-SH (human neuroblastoma epithelial cell) cells and 5 µg of ab217668 [NCI-R159-6]. The resulting DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 10 million reads. The negative IgG control ab172730 is also shown. The University of Geneva owns patents relevant to ChIC (Chromatin Immuno-Cleavage) methods.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-TCF-4 antibody [NCI-R159-6] - BSA and Azide free (ab223073)

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 TCF-4 #2.

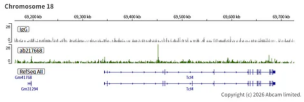
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab217668).



ChIC/CUT&RUN - Anti-TCF-4 antibody [NCI-R159-6] - BSA and Azide free (ab223073)

This data was developed using ab217668, the same antibody clone in a different buffer formulation.

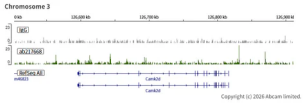
ChIC/CUT&RUN was performed using a pAG-MNase at a final concentration of 700 ng/mL, 2.5 x 10^5 Neuro-2a (mouse neuroblastoma neuroblast) cells and 5 µg of ab217668 [NCI-R159-6]. The resulting DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 10 million reads. The negative IgG control ab172730 is also shown. The University of Geneva owns patents relevant to ChIC (Chromatin Immuno-Cleavage) methods.



ChIC/CUT&RUN sequencing - Anti-TCF-4 antibody [NCI-R159-6] - BSA and Azide free (ab223073)

This data was developed using ab217668, the same antibody clone in a different buffer formulation.

ChIC/CUT&RUN was performed using a pAG-MNase at a final concentration of 700 ng/mL, 2.5 x 10^5 Neuro-2a (mouse neuroblastoma neuroblast) cells and 5 µg of ab217668 [NCI-R159-6]. The resulting DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 10 million reads. The negative IgG control ab172730 is also shown. The University of Geneva owns patents relevant to ChIC (Chromatin Immuno-Cleavage) methods.



ChIC/CUT&RUN - Anti-TCF-4 antibody [NCI-R159-6] - BSA and Azide free (ab223073)

This data was developed using ab217668, the same antibody clone in a different buffer formulation.

ChIC/CUT&RUN was performed using a pAG-MNase at a final concentration of 700 ng/mL, 2.5 x 10^5 Neuro-2a (mouse neuroblastoma neuroblast) cells and 5 µg of ab217668 [NCI-R159-6]. The resulting DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 10 million reads. The negative IgG control ab172730 is also shown. The University of Geneva owns patents relevant to ChIC (Chromatin Immuno-Cleavage) methods.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.