# abcam

# Product datasheet

# Anti-c-Jun antibody [E254] - BSA and Azide free ab218576



## 5 Images

#### Overview

Product name Anti-c-Jun antibody [E254] - BSA and Azide free

**Description** Rabbit monoclonal [E254] to c-Jun - BSA and Azide free

Host species Rabbit

**Specificity** The mouse and rat recommendation is based on the WB results. We do not guarantee IHC-P for

mouse and rat.

Tested applications Suitable for: WB, IHC-P, IP, ChIP

Unsuitable for: Flow Cyt

**Species reactivity** Reacts with: Mouse, Rat, Human

Predicted to work with: Pig ...

**Immunogen** Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

**Epitope** <u>ab32137</u> reacts with an epitope located in the N terminal region of c-Jun.

Positive control WB: NIH 3T3 and HeLa cell lysate. ICC/IF HeLa and NIH/3T3 cells. IHC-P: Human lung carcinoma

tissue IP: HEK-293 and NIH3T3 cells.

**General notes** ab218576 is the carrier-free version of <u>ab32137</u>.

Our <u>carrier-free</u> antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for

increased conjugation efficiency.

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 <u>conjugation kits</u> 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.

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

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

- High batch-to-batch consistency and reproducibility
- Improved sensitivity and specificity

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- Long-term security of supply
- Animal-free production

For more information **see here**.

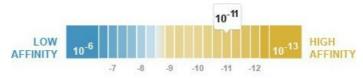
Our RabMAb<sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**<sup>®</sup> **patents**.

# **Properties**

Form Liquid

**Storage instructions** Shipped at 4°C. Store at +4°C. Do Not Freeze.

**Dissociation constant (K<sub>D</sub>)**  $K_D = 2.23 \times 10^{-11} M$ 



Learn more about K<sub>D</sub>

Storage buffer pH: 7.20

Constituent: PBS

Carrier free Yes

Purity Protein A purified

**Clonality** Monoclonal

Clone number E254
Isotype IgG

#### **Applications**

## The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab218576 in the following tested applications.

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

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Application	Abreviews	Notes
WB		Use at an assay dependent concentration. Predicted molecular weight: 36 kDa.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.  The mouse and rat recommendation is based on the WB results. We do not guarantee IHC-P for mouse and rat.
IP		Use at an assay dependent concentration.
ChIP		Use at an assay dependent concentration.

**Application notes** 

Is unsuitable for Flow Cyt.

#### **Target**

#### **Function**

Transcription factor that recognizes and binds to the enhancer heptamer motif 5'-TGA[CG]TCA-3'. Promotes activity of NR5A1 when phosphorylated by HIPK3 leading to increased steroidogenic gene expression upon cAMP signaling pathway stimulation. Involved in activated KRAS-mediated transcriptional activation of USP28 in colorectal cancer (CRC) cells (PubMed:24623306). Binds to the USP28 promoter in colorectal cancer (CRC) cells (PubMed:24623306).

#### Sequence similarities

Belongs to the bZIP family. Jun subfamily.

Contains 1 bZIP (basic-leucine zipper) domain.

# Post-translational modifications

Ubiquitinated by the SCF(FBXW7), leading to its degradation. Ubiquitination takes place following phosphorylation, that promotes interaction with FBXW7.

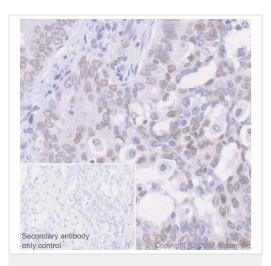
Phosphorylated by CaMK4 and PRKDC; phosphorylation enhances the transcriptional activity. Phosphorylated by HIPK3. Phosphorylated by DYRK2 at Ser-243; this primes the protein for subsequent phosphorylation by GSK3B at Thr-239. Phosphorylated at Thr-239, Ser-243 and Ser-249 by GSK3B; phosphorylation reduces its ability to bind DNA. Phosphorylated by PAK2 at Thr-2, Thr-8, Thr-89, Thr-93 and Thr-286 thereby promoting JUN-mediated cell proliferation and transformation. Phosphorylated by PLK3 following hypoxia or UV irradiation, leading to increase DNA-binding activity.

Acetylated at Lys-271 by EP300.

#### **Cellular localization**

#### Nucleus.

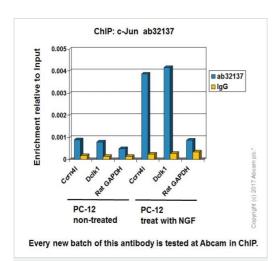
#### **Images**



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-c-Jun antibody [E254] - BSA and Azide free (ab218576)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human lung carcinoma tissue sections labeling c-Jun with purified <u>ab32137</u> at 1:100 dilution (1.2 µg/ml). Heat mediated antigen retrieval was performed using <u>ab93684</u> (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.

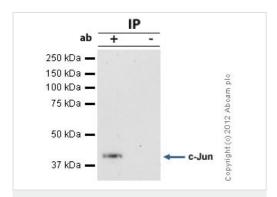
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab32137</u>)



ChIP - Anti-c-Jun antibody [E254] - BSA and Azide free (ab218576)

Chromatin was prepared from PC-12 (starve overnight) + NGF (50 ng/ml 2h) cells according to the Abcam X-ChIP protocol. Cells were fixed with 1% formaldehyde for 10 minutes. The ChIP was performed with 25µg of chromatin, 5µg of ab32137 (blue), and 20µl of protein A/G sepharose beads slurry (10µl of sepharose A beads + 10µl of sepharose G beads). 5µg of rabbit normal IgG was added to the beads control (yellow). The immunoprecipitated DNA was quantified by real time PCR (Sybr green approach).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab32137)



Immunoprecipitation - Anti-c-Jun antibody [E254] - BSA and Azide free (ab218576)

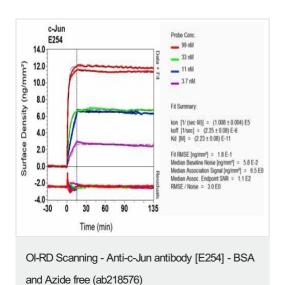
c-Jun was immunoprecipitated using 0.5mg NIH3T3 whole cell extract, 5µg of Rabbit polyclonal to c-Jun and 50µl of protein G magnetic beads (+). No antibody was added to the control (-). The antibody was incubated under agitation with Protein G beads for 10min, NIH3T3 whole cell extract lysate diluted in RIPA buffer was added to each sample and incubated for a further 10min under agitation.

Proteins were eluted by addition of 40µl SDS loading buffer and incubated for 10min at 70°C; 10µl of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with **ab32137**.

Secondary: Mouse monoclonal [SB62a] Secondary Antibody to Rabbit IgG light chain (HRP) (ab99697).

Band: 45kDa; c-Jun

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab32137).



Equilibrium disassociation constant ( $K_D$ ) Learn more about  $K_D$ 

### Click here to learn more about KD

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab32137).



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