abcam

Product datasheet

Anti-MEF2C antibody [EPR19089-202] - ChIP Grade ab211493



★★★★★ 5 Abreviews 7 References 画像数 12

製品の概要

製品名 Anti-MEF2C antibody [EPR19089-202] - ChIP Grade

製品の詳細 Rabbit monoclonal [EPR19089-202] to MEF2C - ChIP Grade

由来種 Rabbit

アプリケーション 適用あり: Flow Cyt (Intra), ChIP, ICC/IF, IHC-P, WB

種交差性 交差種: Mouse, Rat, Human

交差が予測される動物種: Common marmoset 4

Recombinant fragment. This information is proprietary to Abcam and/or its suppliers. 免疫原

ポジティブ・コントロール WB: Human MEF2C recombinant protein; Raji, Jurkat, Ramos, Daudi, RAW264.7, NIH/3T3,C6,

> Wild-type THP-1 and Daudi whole cell lysates; mouse and rat brain lysate. IHC-P: Human skeletal muscle and endometrial carcinoma tissue; Mouse and rat spleen tissue. ICC/IF: Raji cells. Flow:

Jurkat cells. ChIP: Chromatin prepared from HUVEC cells.

特記事項 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**.

製品の特性

製品の状態 Liquid

保存方法 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.

バッファー pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: PBS, 40% Glycerol, 0.05% BSA

精製度 Protein A purified ポリモノ モノクローナル

クローン名

EPR19089-202

アイソタイプ

lgG

アプリケーション

The Abpromise guarantee Abpromise保証は、次のテスト済みアプリケーションにおけるab211493の使用に適用されますアプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
Flow Cyt (Intra)		1/500.
ChIP		Use 5 µg for 25 µg of chromatin.
ICC/IF		1/500.
IHC-P	★★★★★ (4)	Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. Use at 1:500 (human), 1:1000 (mouse, rat) dilution.
WB		1/1000. Predicted molecular weight: 51 kDa.

ターゲット情報

機能

Transcription activator which binds specifically to the MEF2 element present in the regulatory regions of many muscle-specific genes. Controls cardiac morphogenesis and myogenesis, and is also involved in vascular development. Plays an essential role in hippocampal-dependent learning and memory by suppressing the number of excitatory synapses and thus regulating basal and evoked synaptic transmission. Crucial for normal neuronal development, distribution, and electrical activity in the neocortex. Necessary for proper development of megakaryocytes and platelets and for bone marrow B lymphopoiesis. Required for B-cell survival and proliferation in response to BCR stimulation, efficient lgG1 antibody responses to T-cell-dependent antigens and for normal induction of germinal center B cells. May also be involved in neurogenesis and in the development of cortical architecture (By similarity). Isoform 3 and isoform 4, which lack the repressor domain, are more active than isoform 1 and isoform 2.

組織特異性

関連疾患

Expressed in brain and skeletal muscle.

Defects in MEF2C are the cause of mental retardation-stereotypic movements-epilepsy and/or cerebral malformations (MRSME) [MIM:613443]. It is a disorder characterized by severe mental retardation, absent speech, hypotonia, poor eye contact and stereotypic movements. Dysmorphic features include high broad forehead with variable small chin, short nose with anteverted nares, large open mouth, upslanted palpebral fissures and prominent eyebrows. Some patients have seizures.

配列類似性

Belongs to the MEF2 family.

Contains 1 MADS-box domain.

Contains 1 Mef2-type DNA-binding domain.

発生段階

Expression is highest during the early stages of postnatal development, at later stages levels

greatly decrease.

ドメイン

The beta domain, missing in a number of isoforms, is required for enhancement of transcriptional activity.

翻訳後修飾

Phosphorylation on Ser-59 enhances DNA binding activity (By similarity). Phosphorylation on Ser-396 is required for Lys-391 sumoylation and inhibits transcriptional activity.

Acetylated by p300 on several sites in diffentiating myocytes. Acetylation on Lys-4 increases DNA binding and transactivation.

Sumoylated on Lys-391 by SUMO2 but not by SUMO1 represses transcriptional activity. Proteolytically cleaved in cerebellar granule neurons, probably by caspase 7, following

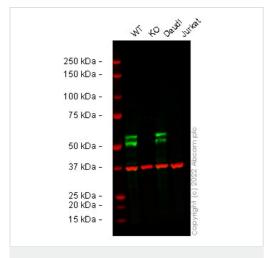
neurotoxicity. Preferentially cleaves the CDK5-mediated hyperphosphorylated form which leads to

neuron apoptosis and transcriptional inactivation.

細胞内局在

Nucleus.

画像



Western blot - Anti-MEF2C antibody [EPR19089-202] - ChIP Grade (ab211493) **All lanes :** Anti-MEF2C antibody [EPR19089-202] - ChIP Grade (ab211493) at 1/1000 dilution

Lane 1: Wild-type THP-1 cell lysate

Lane 2: MEF2C knockout THP-1 cell lysate

Lane 3 : Daudi cell lysate

Lane 4 : Jurkat cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution

Performed under reducing conditions.

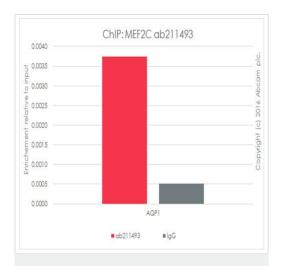
Predicted band size: 51 kDa **Observed band size:** 55/60 kDa

False colour image of Western blot: Anti-MEF2C antibody [EPR19089-202] - ChIP Grade staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (ab8245) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab211493 was shown to bind specifically to MEF2C. A band was observed at 55/60 kDa in wild-type THP-1 cell lysates with no signal observed at this size in MEF2C knockout cell line. To generate this image, wild-type and MEF2C knockout THP-1 cell lysates were

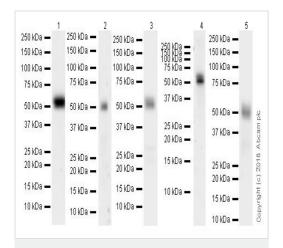
analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween[®] 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit lgG H&L 800CW and Goat anti-Mouse lgG H&L 680RD at 1/20000 dilution.

Chromatin was prepared from HUVEC cells according to the Abcam X-ChIP protocol. Cells were fixed with formaldehyde for 10 minutes. The ChIP was performed with: 25 μg of chromatin, 5 μg of ab211493 (red), and 20 μl of protein A/G sepharose beads slurry (10 μl of sepharose A beads + 10 μl of sepharose G beads). 5 μg of rabbit normal IgG was added to the beads control (grey). The immunoprecipitated DNA was quantified by real time PCR (SYBR green chemistry).

ChIP was performed according to the literature (PMID: 26923194).



ChIP - Anti-MEF2C antibody [EPR19089-202] - ChIP Grade (ab211493)



Western blot - Anti-MEF2C antibody [EPR19089-202] - ChIP Grade (ab211493)

All lanes : Anti-MEF2C antibody [EPR19089-202] - ChIP Grade (ab211493) at 1/1000 dilution

Lane 1 : RAW264.7 (mouse Abelson murine leukemia virus-induced tumor macrophage), whole cell lysate

Lane 2: NIH/3T3 (mouse embryonic fibroblast), whole cell lysate

Lane 3: C6 (rat glial tumor glial cell), whole cell lysate

Lane 4 : Mouse brain lysate

Lane 5: Rat brain lysate

Lysates/proteins at 10 µg per lane.

Secondary

Lanes 1-3 & 5 : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution (Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated)

Lane 4 : Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/50000 dilution (Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated)

Predicted band size: 51 kDa

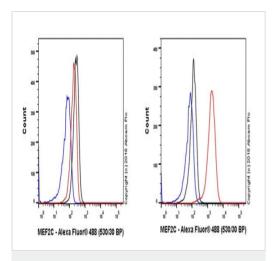
Observed band size: 50-60 kDa

Blocking and diluting buffer and concentration: 5% NFDM/TBST Exposure time:

Lanes 1-2,4:5 minutes

Lane 3: 15 seconds

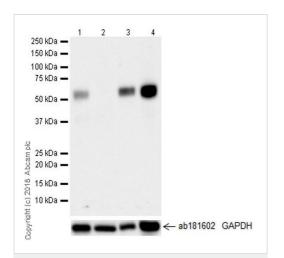
Lane 5: 26 seconds



Flow Cytometry (Intracellular) - Anti-MEF2C antibody [EPR19089-202] - ChIP Grade (ab211493)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol permeabilsed Jurkat (Human T cell leukemia T lymphocyte, Left) / Raji (Human Burkitt's lymphoma B lymphocyte, Right) cell lines labelling MEF2C with ab211493 at 1/500 dilution (Red) compared with the isotype control Rabbit monoclonal lgG (ab172730) (Black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). Goat anti rabbit lgG Alexa Fluor® 488 (ab150077), at 1/2000 dilution was used as the secondary antibody.

Negative control: Jurkat (PMID: 27876533).



Western blot - Anti-MEF2C antibody [EPR19089-202] - ChIP Grade (ab211493)

All lanes : Anti-MEF2C antibody [EPR19089-202] - ChIP Grade (ab211493) at 1/1000 dilution

Lane 1 : Raji (human Burkitt's lymphoma B lymphocyte), whole cell lysate 20 µg

Lane 2 : Jurkat (human T cell leukemia T lymphocyte), whole cell lysate 20 μ g

Lane 3 : Ramos (human Burkitt's lymphoma B lymphocyte), whole cell lysate 20µg

Lane 4 : Daudi (human Burkitt's lymphoma lymphoblast), whole cell lysate 20 µg

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/50000 dilution (Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated)

Predicted band size: 51 kDa **Observed band size:** 50-60 kDa

Blocking and diluting buffer and concentration: 5% NFDM/TBST Exposure time: 92 seconds.

The expression profile observed is consistent with the literature (PMID: 18450586). **Negative control**: Jurkat (PMID: 27876533)

All lanes : Anti-MEF2C antibody [EPR19089-202] - ChIP Grade (ab211493) at 1/1000 dilution

Lane 1 : Human MEF2C recombinant protein (aa271-493) 10 ng
Lane 2 : Human MEF2A recombinant protein (aa1-499,

ab204772) 10 ng

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution (Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated)

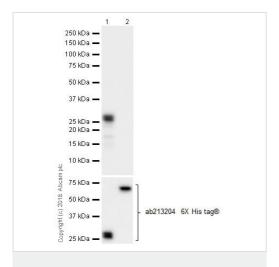
Predicted band size: 51 kDa
Observed band size: 26 kDa

Blocking and diluting buffer and concentration: 5% NFDM/TBST Exposure time: 3 seconds.

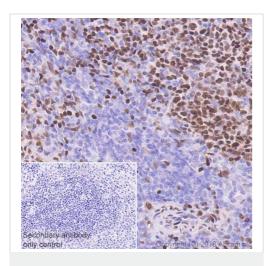
Immunohistochemical analysis of paraffin-embedded rat spleen tissue labelling MEF2C with ab211493 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) ready to use. Nuclear staining in B lymphocytes of rat spleen but not in T cells in the periarterial lymphatic sheath is observed (PMID: 8506376, PMID 15703219). Counterstained with hematoxylin.

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

Perform heat mediated antigen retrieval with EDTA buffer pH 9 before commencing with IHC staining protocol.

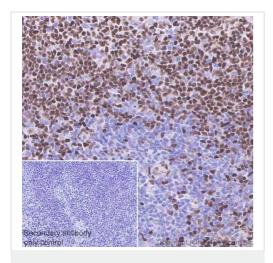


Western blot - Anti-MEF2C antibody [EPR19089-202] - ChIP Grade (ab211493)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-MEF2C antibody

[EPR19089-202] - ChIP Grade (ab211493)

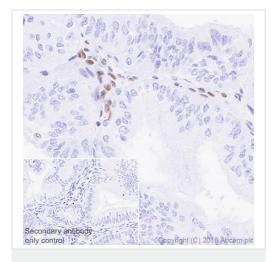


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-MEF2C antibody

[EPR19089-202] - ChIP Grade (ab211493)

Immunohistochemical analysis of paraffin-embedded mouse spleen tissue labelling MEF2C with ab211493 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) ready to use. Nuclear staining in B lymphocytes of mouse spleen but not in T cells in the periarterial lymphatic sheath is observed (PMID: 8506376, PMID 15703219). Counterstained with hematoxylin. Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) ready to use.

Perform heat mediated antigen retrieval with EDTA buffer pH 9 before commencing with IHC staining protocol.

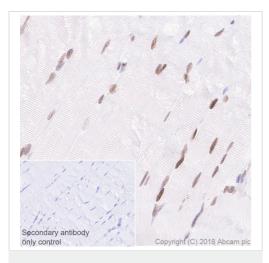


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-MEF2C antibody

[EPR19089-202] - ChIP Grade (ab211493)

Immunohistochemical analysis of paraffin-embedded human endometrial carcinoma tissue labelling MEF2C with ab211493 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) ready to use. Nuclear staining in leukocytes but not in tumor cells of human endometrial carcinoma is observed (PMID: 8506376, PMID 15703219). Counterstained with hematoxylin. Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) ready to use.

Perform heat mediated antigen retrieval with EDTA buffer pH 9 before commencing with IHC staining protocol.

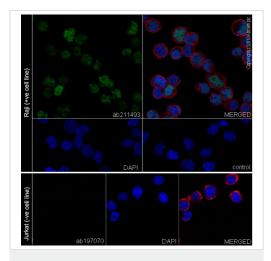


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-MEF2C antibody

[EPR19089-202] - ChIP Grade (ab211493)

Immunohistochemical analysis of paraffin-embedded human skeletal muscle tissue labelling MEF2C with ab211493 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) ready to use. Nuclear staining in human skeletal muscle cells is observed(PMID: 8506376, PMID 15703219). Counterstained with hematoxylin. Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) ready to use.

Perform heat mediated antigen retrieval with EDTA buffer pH 9 before commencing with IHC staining protocol.

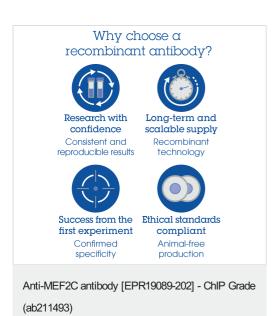


Immunocytochemistry/ Immunofluorescence - Anti-MEF2C antibody [EPR19089-202] - ChIP Grade (ab211493)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% Triton X-100 permeabilised Raji (human Burkitt's lymphoma B lymphocyte) cells labelling MEF2C with ab211493 at 1/500 dilution, followed by AlexaFluor®488 Goat anti-Rabbit secondary (ab150077) at 1/1000 dilution (green). Confocal image showing nuclear staining in Raji cell line. Negative control: Jurkat (PMID: 27876533). DAPI was used as the nuclear counterstain, and the Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) (ab195889 antibody was used as a counterstain at 1/200 dilution.

The negetive controls are as follows:

- -ve control 1: <u>ab197070</u> on jurkat (human T cell leukemia cell line from peripheral blood) cells.
- -ve control 2: Jurkat cells stained with DAPI.
- -ve control 3: Merged negetive contol images.



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