Product datasheet

Anti-Argonaute-2 antibody [2E12-1C9] - ChIP Grade
ab57113

Overview

Product name
Anti-Argonaute-2 antibody [2E12-1C9] - ChIP Grade

Description
Mouse monoclonal [2E12-1C9] to Argonaute-2 - ChIP Grade

Host species
Mouse

Tested applications
Suitable for: CHIPseq, WB, ICC/IF, IHC-P, IP, Flow Cyt, ChIP

Species reactivity
Reacts with: Chicken, Human, Zebrafish
Does not react with: Xenopus laevis

Immunogen
Recombinant fragment corresponding to Human Argonaute-2 aa 483-859.

General notes
This product was changed from ascites to tissue culture supernatant on 30th April 2019. Please note that the dilutions may need to be adjusted accordingly. If you have any questions, please do not hesitate to contact our scientific support team.

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 or -80°C. Avoid freeze / thaw cycle.

Storage buffer
pH: 7.20
Constituent: PBS

Purity
Tissue culture supernatant

Clonality
Monoclonal

Clone number
2E12-1C9

Isotype
IgG1

Light chain type
kappa

Applications

Our Abpromise guarantee covers the use of ab57113 in the following tested applications.

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

Required for RNA-mediated gene silencing (RNAi) by the RNA-induced silencing complex (RISC). The 'minimal RISC' appears to include EIF2C2/AGO2 bound to a short guide RNA such as a microRNA (miRNA) or short interfering RNA (siRNA). These guide RNAs direct RISC to complementary mRNAs that are targets for RISC-mediated gene silencing. The precise mechanism of gene silencing depends on the degree of complementarity between the miRNA or siRNA and its target. Binding of RISC to a perfectly complementary mRNA generally results in silencing due to endonucleolytic cleavage of the mRNA specifically by EIF2C2/AGO2. Binding of RISC to a partially complementary mRNA results in silencing through inhibition of translation, and this is independent of endonuclease activity. May inhibit translation initiation by binding to the 7-methylguanosine cap, thereby preventing the recruitment of the translation initiation factor eIF4-E. May also inhibit translation initiation via interaction with EIF6, which itself binds to the 60S ribosomal subunit and prevents its association with the 40S ribosomal subunit. The inhibition of translational initiation leads to the accumulation of the affected mRNA in cytoplasmic processing bodies (P-bodies), where mRNA degradation may subsequently occur. In some cases RISC-mediated translational repression is also observed for miRNAs that perfectly match the 3' untranslated region (3'-UTR). Can also upregulate the translation of specific mRNAs under certain growth conditions. Binds to the AU element of the 3'-UTR of the TNF (TNF-alpha) mRNA and upregulates translation under conditions of serum starvation. Also required for transcriptional gene silencing (TGS), in which short RNAs known as antigenic RNAs or agRNAs direct the transcriptional repression of complementary promoter regions.

**Sequence similarities**

Belongs to the argonaute family. Ago subfamily.
Contains 1 PAZ domain.
Contains 1 Piwi domain.

**Domain**

The Piwi domain may perform RNA cleavage by a mechanism similar to that of RNase H. However while RNase H utilizes a triad of Asp-Asp-Glu (DDE) for metal ion coordination, this protein appears to utilize a triad of Asp-Asp-His (DDH).

**Post-translational modifications**

Hydroxylated. 4-hydroxylation appears to enhance protein stability but is not required for miRNA-binding or endonuclease activity.

**Cellular localization**

Cytoplasm > P-body. Nucleus. Translational repression of mRNAs results in their recruitment to P-bodies. Translocation to the nucleus requires IMP8.
Western blot - Anti-Argonaute-2 antibody [2E12-1C9] - ChIP Grade (ab57113) + MCF7 cell lysate

**Predicted band size:** 97 kDa

**Observed band size:** 97 kDa

Ago2 / eIF2C2 antibody (ab57113) used in immunofluorescence at 10μg/ml on HeLa cells.

This image was generated using the ascites version of the product.

Ago2 / eIF2C2 antibody (ab57113) used in immunohistochemistry at 3μg/ml on formalin fixed and paraffin embedded human stomach.

This image was generated using the ascites version of the product.
Overlay histogram showing HeLa cells stained with ab57113 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab57113, 1µg/1x10⁶ cells) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-mouse IgG (H+L) (ab96879) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was mouse IgG1 [ICIGG1] (ab91353, 2µg/1x10⁶ cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in HeLa cells fixed with 4% paraformaldehyde/permeabilized in 0.1% PBS-Tween used under the same conditions.

This image was generated using the ascites version of the product.

**All lanes**: Anti-Argonaute-2 antibody [2E12-1C9] - ChIP Grade (ab57113) at 5 µg/ml

**Lane 1**: Argonaute-2 overexpressing HEK-293 cells line
**Lane 2**: Argonaute-2 overexpressing HEK-293 cell line cotransfected with Argonaute-2 validated chimera RNAi.

**Predicted band size**: 97 kDa
**Observed band size**: 42 kDa

**why is the actual band size different from the predicted?**

The band at about 42 kDa corresponds to the recombinant fragment of human Argonaute-2 aa 483-859 (377 aa length).

The loading control is GAPDH.

This image was generated using the ascites version of the product.
Western blot detection against the recombinant fragment immunogen (68 KDa for a.a. ~377 plus GST tag +26 kDa).

This image was generated using the ascites version of the product.

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