Product datasheet

Anti-Argonaute-2 antibody ab5072

Overview

Product name
Anti-Argonaute-2 antibody

Description
Rabbit polyclonal to Argonaute-2

Host species
Rabbit

Tested applications
Suitable for: ICC/IF, WB

Species reactivity
React with: Drosophila melanogaster
Does not react with: Human

Immunogen
Synthetic peptide corresponding to Drosophila melanogaster Argonaute-2 aa 550-650 conjugated to keyhole limpet haemocyanin.
(Peptide available as ab24177)

Positive control
This antibody gave a positive result in WB on Drosophila lysate.

General notes
Please note the immunogen for this antibody has no sequence alignment to human dIFC2. When tested in human, non-specific binding is observed.

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
Preservative: 0.02% Sodium Azide
Constituents: 1% BSA, PBS, pH 7.4

Purity
Immunogen affinity purified

Clonality
Polyclonal

Isotype
IgG

Applications

Our Abpromise guarantee covers the use of ab5072 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 antigene 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.

Images
Anti-Argonaute-2 antibody (ab5072) at 1 µg/ml + Schneider L2 whole cell lysate (ab14893) at 20 µg

**Secondary**

Goat Anti-Rabbit IgG H&L (HRP) at 1/50000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

**Predicted band size:** 106 kDa

**Observed band size:** 106 kDa

**Additional bands at:** 120 kDa, 19 kDa, 50 kDa, 65 kDa. We are unsure as to the identity of these extra bands.

**Exposure time:** 30 seconds

This blot was produced using a 4-12% Bis-tris gel under the MOPS buffer system. The gel was run at 200V for 50 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using 2% Bovine Serum Albumin before being incubated with ab5072 overnight at 4°C. Antibody binding was detected using an anti-rabbit antibody conjugated to HRP, and visualised using ECL development solution ab133406.

ab5072 staining Ago2 / eIF2C2 (green) in fruit fly (Drosophila melanogaster) OSS cells by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with formaldehyde, permeabilized with 0.2% Tween and blocked with 1% BSA for 1 hour at 22°C. Samples were incubated with primary antibody (1/200) for 16 hours at 4°C. An Alexa Fluor® 488-conjugated donkey anti-rabbit IgG monoclonal (1/5000) was used as the secondary antibody.

**ab24609** was used to stain Nuclear Pore Complex Proteins (red).
Anti-Argonaute-2 antibody (ab5072) at 1/500 dilution + Drosophila lysate at 20 µg

**Secondary**
Goat Anti-Rabbit IgG H&L (HRP) (ab6721)

**Predicted band size:** 106 kDa
**Observed band size:** 55,95 kDa

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

Rabbit polyclonal to Ago2 (ab5072) at 1/500 on Drosophila lysate (20ug).

Detects a band of approximately 55 + 95 kDa (predicted molecular weight: 106 kDa). Swissprot suggests Ago 2 may have 50kDa form.

Secondary antibody - Goat polyclonal to rabbit IgG (HRP) - ab6721. Swissprot suggests Ago 2 may have 50kDa form.

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

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