abcam

Product datasheet

Anti-beta Arrestin 1 antibody [E274] ab32099



重组 RabMAb

★★★★ 4 Abreviews 42 References 12 图像

概述

产品名称 Anti-beta Arrestin 1抗体[E274]

描述 兔单克隆抗体[E274] to beta Arrestin 1

宿主 Rabbit

特异性 The antibody immunogen shares 90% homology with ARRB2 (P32121) which has similar MW

than ARRB1. Therefore, it is likely that the antbody will cross-react with ARRB2. However, we

haven't performed any experiment with recombinant protein to confirm this.

经测试应用 适用于: Flow Cyt (Intra), ICC/IF, WB, IHC-P

种属反应性 与反应: Mouse, Rat, Human

预测可用于: Cow 🕰

免疫原 Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

阳性对照 WB: HEK293, Jurkat, SH-SY5Y, C2C12 and PC-12 cell lysates. IHC-P: Human lymph node,

human breast carcinoma, mouse cerebral cortex and rat cerebral cortex tissues, ICC/IF: PC-3

cells.

常规说明 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**.

性能

形式 Liquid

存放说明 Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Avoid freeze / thaw cycle.

存储溶液 pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol, 0.05% BSA

纯**度** Protein A purified

 克隆
 单克隆

 克隆编号
 E274

 同种型
 IqG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab32099于以下的经测试应用

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
Flow Cyt (Intra)		1/10 - 1/20. ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
ICC/IF	★★★★ <u>(2)</u>	1/100 - 1/150.
WB	*** <u>*</u>	1/500 - 1/1000. Predicted molecular weight: 50 kDa.
IHC-P	★★★★ (1)	1/100 - 1/250. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See IHC antigen retrieval protocols.

靶标

功能

Functions in regulating agonist-mediated G-protein coupled receptor (GPCR) signaling by mediating both receptor desensitization and resensitization processes. During homologous desensitization, beta-arrestins bind to the GPRK-phosphorylated receptor and sterically preclude its coupling to the cognate G-protein; the binding appears to require additional receptor determinants exposed only in the active receptor conformation. The beta-arrestins target many receptors for internalization by acting as endocytic adapters (CLASPs, clathrin-associated sorting proteins) and recruiting the GPRCs to the adapter protein 2 complex 2 (AP-2) in clathrin-coated pits (CCPs). However, the extent of beta-arrestin involvement appears to vary significantly depending on the receptor, agonist and cell type. Internalized arrestin-receptor complexes traffic to intracellular endosomes, where they remain uncoupled from G-proteins. Two different modes of arrestin-mediated internalization occur. Class A receptors, like ADRB2, OPRM1, ENDRA, D1AR and ADRA1B dissociate from beta-arrestin at or near the plasma membrane and undergo rapid recycling. Class B receptors, like AVPR2, AGTR1, NTSR1, TRHR and TACR1 internalize as a complex with arrestin and traffic with it to endosomal vesicles, presumably as desensitized receptors, for extended periods of time. Receptor resensitization then requires that receptorbound arrestin is removed so that the receptor can be dephosphorylated and returned to the plasma membrane. Involved in internalization of P2RY4 and UTP-stimulated internalization of P2RY2. Involved in phopshorylation-dependent internalization of OPRD1 ands subsequent recycling. Involved in the degradation of cAMP by recruiting cAMP phosphodiesterases to ligandactivated receptors. Beta-arrestins function as multivalent adapter proteins that can switch the GPCR from a G-protein signaling mode that transmits short-lived signals from the plasma membrane via small molecule second messengers and ion channels to a beta-arrestin signaling

mode that transmits a distinct set of signals that are initiated as the receptor internalizes and transits the intracellular compartment. Acts as signaling scaffold for MAPK pathways such as MAPK1/3 (ERK1/2). ERK1/2 activated by the beta-arrestin scaffold is largely excluded from the nucleus and confined to cytoplasmic locations such as endocytic vesicles, also called betaarrestin signalosomes. Recruits c-Src/SRC to ADRB2 resulting in ERK activation. GPCRs for which the beta-arrestin-mediated signaling relies on both ARRB1 and ARRB2 (codependent regulation) include ADRB2, F2RL1 and PTH1R. For some GPCRs the beta-arrestin-mediated signaling relies on either ARRB1 or ARRB2 and is inhibited by the other respective beta-arrestin form (reciprocal regulation). Inhibits ERK1/2 signaling in AGTR1- and AVPR2-mediated activation (reciprocal regulation). Is required for SP-stimulated endocytosis of NK1R and recruits c-Src/SRC to internalized NK1R resulting in ERK1/2 activation, which is required for the antiapoptotic effects of SP. Is involved in proteinase-activated F2RL1-mediated ERK activity. Acts as signaling scaffold for the AKT1 pathway. Is involved in alpha-thrombin-stimulated AKT1 signaling. Is involved in IGF1-stimulated AKT1 signaling leading to increased protection from apoptosis. Involved in activation of the p38 MAPK signaling pathway and in actin bundle formation. Involved in F2RL1-mediated cytoskeletal rearrangement and chemotaxis. Involved in AGTR1-mediated stress fiber formation by acting together with GNAQ to activate RHOA. Appears to function as signaling scaffold involved in regulation of MIP-1-beta-stimulated CCR5dependent chemotaxis. Involved in attenuation of NF-kappa-B-dependent transcription in response to GPCR or cytokine stimulation by interacting with and stabilizing CHUK. May serve as nuclear messenger for GPCRs. Involved in OPRD1-stimulated transcriptional regulation by translocating to CDKN1B and FOS promoter regions and recruiting EP300 resulting in acetylation of histone H4. Involved in regulation of LEF1 transcriptional activity via interaction with DVL1 and/or DVL2 Also involved in regulation of receptors others than GPCRs. Involved in Tolllike receptor and IL-1 receptor signaling through the interaction with TRAF6 which prevents TRAF6 autoubiquitination and oligomerization required for activation of NF-kappa-B and JUN. Binds phosphoinositides. Binds inositolhexakisphosphate (InsP6).

序列相似性

结构域

翻译后修饰

细胞定位

Belongs to the arrestin family.

The [DE]-X(1,2)-F-X-X-[FL]-X-X-R motif mediates interaction the AP-2 complex subunit AP2B1 (By similarity). Binding to phosphorylated GPCRs induces a conformationanl change that exposes the motif to the surface.

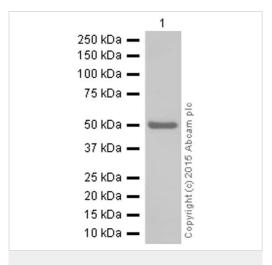
The N-terminus binds InsP6 with low affinity.

The C-terminus binds InsP6 with high affinity.

Constitutively phosphorylated at Ser-412 in the cytoplasm. At the plasma membrane, is rapidly dephosphorylated, a process that is required for clathrin binding and ADRB2 endocytosis but not for ADRB2 binding and desensitization. Once internalized, is rephosphorylated.

The ubiquitination status appears to regulate the formation and trafficking of beta-arrestin-GPCR complexes and signaling. Ubiquitination appears to occur GPCR-specific. Ubiquitinated by MDM2; the ubiquitination is required for rapid internalization of ADRB2. Deubiquitinated by USP33; the deubiquitination leads to a dissociation of the beta-arrestin-GPCR complex. Stimulation of a class A GPCR, such as ADRB2, induces transient ubiquitination and subsequently promotes association with USP33.

Cytoplasm. Nucleus. Cell membrane. Membrane > clathrin-coated pit. Cell projection > pseudopodium. Cytoplasmic vesicle. Translocates to the plasma membrane and colocalizes with antagonist-stimulated GPCRs. The monomeric form is predominantly located in the nucleus. The oligomeric form is located in the cytoplasm. Translocates to the nucleus upon stimulation of OPRD1.



Western blot - Anti-beta Arrestin 1 antibody [E274] (ab32099)

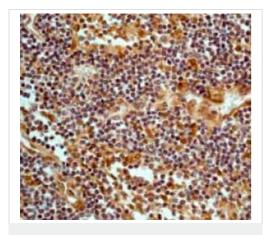
Anti-beta Arrestin 1 antibody [E274] (ab32099) at 1/1000 dilution (purified) + Jurkat whole cell lysate at 20 µg

Secondary

Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 50 kDa **Observed band size:** 50 kDa

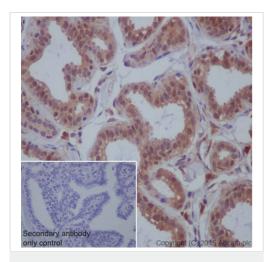
Blocking and dilution buffer: 5% NFDM/TBST



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-beta Arrestin 1 antibody [E274] (ab32099)

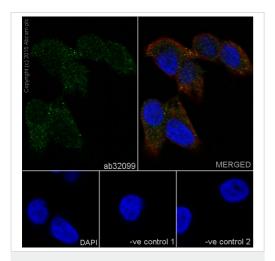
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human lymph node tissue labelling beta Arrestin 1 with unpurified ab32099 at a dilution of 1/100.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-beta Arrestin 1 antibody [E274] (ab32099)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human breast carcinoma tissue labelling beta Arrestin 1 with purified ab32099 at a dilution of 1/250. Heat mediated antigen retrieval was performed using EDTA buffer pH 9. ab97051, a HRP-conjugated goat anti-rabbit lgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.

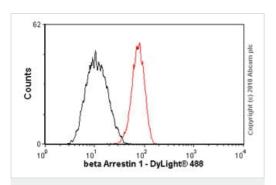


Immunocytochemistry/ Immunofluorescence - Antibeta Arrestin 1 antibody [E274] (ab32099)

Immunocytochemistry/Immunofluorescence analysis of PC-3 cells labelling beta Arrestin 1 with purified ab32099 at a dilution of 1/150. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. ab150077, an Alexa Fluor 488-conjugated goat anti-rabbit IgG (1/1000) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain. ab7291, a mouse anti-tubulin (1/1000) and ab150120, an Alexa Fluor 594-conjugated goat anti-mouse IgG (1/1000) were also used.

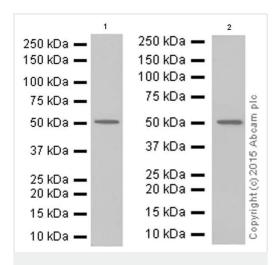
Control 1: primary antibody (1/150) and secondary antibody, **ab150120**, an Alexa Fluor[®] 594-conjugated goat anti-mouse IgG (1/1000).

Control 2: <u>ab7291</u> (1/1000) and secondary antibody, <u>ab150077</u>, an Alexa Fluor[®] 488-conjugated goat anti-rabbit lgG (1/1000).



Flow Cytometry (Intracellular) - Anti-beta Arrestin 1 antibody [E274] (ab32099)

Overlay histogram showing PC3 cells stained with unpurified ab32099 (red line). The cells were fixed with 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 (unpurified ab32099, 1/10 dilution) for 30 min at 22°C. The secondary antibody used was DyLight 488 goat anti-rabbit lgG (H+L) (ab96899) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit monoclonal lgG (1µg/1x106 cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a decreased signal in PC3 cells fixed with 4% paraformaldehyde (10 min)/permeabilized in 0.1% PBS-Tween used under the same conditions.



Western blot - Anti-beta Arrestin 1 antibody [E274] (ab32099)

All lanes : Anti-beta Arrestin 1 antibody [E274] (ab32099) at 1/2000 dilution (purified)

Lane 1: HEK293 whole cell lysate

Lane 2: SH-SY5Y whole cell lysate

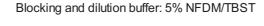
Lysates/proteins at 10 µg per lane.

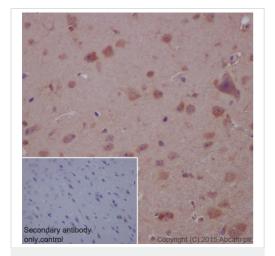
Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) ($\underline{ab97051}$) at 1/20000

dilution

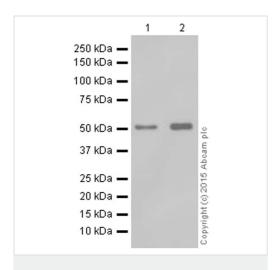
Predicted band size: 50 kDa **Observed band size:** 50 kDa





Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-beta Arrestin 1 antibody [E274] (ab32099)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse cerebral cortex tissue labelling beta Arrestin 1 with purified ab32099 at a dilution of 1/250. Heat mediated antigen retrieval was performed using EDTA buffer pH 9. ab97051, a HRP-conjugated goat anti-rabbit lgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.



Western blot - Anti-beta Arrestin 1 antibody [E274] (ab32099)

All lanes : Anti-beta Arrestin 1 antibody [E274] (ab32099) at 1/1000 dilution (purified)

Lane 1 : C2C12 whole cell lysate

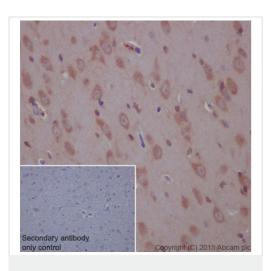
Lane 2 : PC-12 whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

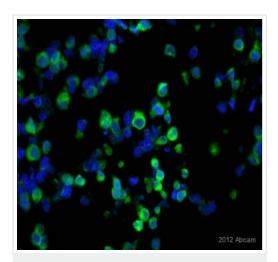
Predicted band size: 50 kDa Observed band size: 50 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-beta Arrestin 1 antibody [E274] (ab32099)

Blocking and dilution buffer: 5% NFDM/TBST

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of rat cerebral cortex tissue labelling beta Arrestin 1 with purified ab32099 at a dilution of 1/250. Heat mediated antigen retrieval was performed using EDTA buffer pH 9. ab97051, a HRP-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.



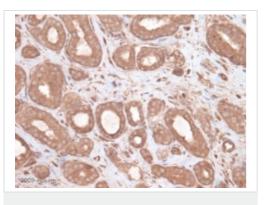
Immunocytochemistry/ Immunofluorescence - Antibeta Arrestin 1 antibody [E274] (ab32099)

This image is courtesy of an anonymous Abreview

Unpurified ab32099 staining beta Arrestin 1 in C4-2B (Human prostate cancer cell line) by ICC/IF

(Immunocytochemistry/immunofluorescence).

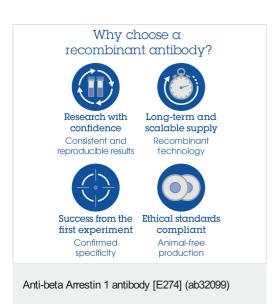
Cells were fixed with paraformaldehyde and blocked with 1% serum for 1 hour at 21°C. Samples were incubated with primary antibody (1/100 in diluent) for 1 hour at 21°C. An Alexa Fluor® 488conjugated goat anti-rabbit polyclonal IgG (1/200) was used as the secondary antibody.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-beta Arrestin 1 antibody [E274] (ab32099)

This image is a courtesy of Anonymous Abreview

Unpurified ab32099 staining beta Arrestin 1 in human prostate carcinoma tissue section by Immunohistochemistry (Formalin/PFAfixed paraffin-embedded sections). Tissue underwent paraformaldehyde fixation before heat mediated antigen retrieval in Tris/EDTA pH9.0 and then blocking with 1% donkey serum for 1 hour at 20°C was performed. The primary antibody was diluted 1/100 and incubated with sample for 1 hour at 20°C in PBS. A Biotin conjugated donkey polyclonal to rabbit IgG was used undiluted as secondary antibody.



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