abcam

ab232516

Anti-ROR gamma antibody [EPR20006] - BSA and Azide free

Recombinant | RabMAb

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	EPR20006
Purification technique	Affinity purification Protein A
Specificity	Our data suggests that this antibody is specific to ROR gamma isoform 2 which is known as ROR gammaT.
Concentration	1.088 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Mouse
Dilution info	-
Notes	-

WB

Expected

Species	Mouse
Dilution info	Use at an assay dependent concentration.
Notes	-

IHC-P

Tested

Species	Mouse
Dilution info	-
Notes	Antigen Retrieval Buffer (100X Tris-EDTA Buffer, pH 9.0) (ab93684).

Flow Cyt (Intra)

Tested

Species	Mouse
Dilution info	-
Notes	-

Target data

See full target information Rorc

Function Nuclear receptor that binds DNA as a monomer to ROR response elements (RORE) containing a single core motif half-site 5'-AGGTCA-3' preceded by a short A-T-rich sequence. Key regulator of cellular differentiation, immunity, peripheral circadian rhythm as well as lipid, steroid, xenobiotics and glucose metabolism. Considered to have intrinsic transcriptional activity, have some natural ligands like oxysterols that act as agonists (25-hydroxycholesterol) or inverse agonists (7-oxygenated sterols), enhancing or repressing the transcriptional activity, respectively. Recruits distinct combinations of cofactors to target gene regulatory regions to modulate

their transcriptional expression, depending on the tissue, time and promoter contexts (PubMed:17666523, PubMed:19381306, PubMed:19965867, PubMed:21853531, PubMed:22789990, PubMed:23723244). Regulates the circadian expression of clock genes such as CRY1, BMAL1 and NR1D1 in peripheral tissues and in a tissue-selective manner (PubMed:22753030). Competes with NR1D1 for binding to their shared DNA response element on some clock genes such as BMAL1, CRY1 and NR1D1 itself, resulting in NR1D1-mediated repression or RORC-mediated activation of the expression, leading to the circadian pattern of clock genes expression. Therefore influences the period length and stability of the clock (PubMed:22753030). Involved in the regulation of the rhythmic expression of genes involved in glucose and lipid metabolism, including PLIN2 and AVPR1A. Negative regulator of adipocyte differentiation through the regulation of early phase genes expression, such as MMP3. Controls adipogenesis as well as adipocyte size and modulates insulin sensitivity in obesity. In liver, has specific and redundant functions with RORA as positive or negative modulator of expression of genes encoding phase I and Phase II proteins involved in the metabolism of lipids, steroids and xenobiotics, such as SULT1E1 (PubMed:21853531). Also plays a role in the regulation of hepatocyte glucose metabolism through the regulation of G6PC1 and PCK1. Regulates the rhythmic expression of PROX1 and promotes its nuclear localization. Plays an indispensable role in the induction of IFN-gamma dependent anti-mycobacterial systemic immunity (By similarity). Isoform 2. Nuclear receptor essential for thymopoiesis and the development of several secondary lymphoid tissues, including lymph nodes and Peyer's patches (PubMed:10602018, PubMed:14691482, PubMed:16148126, PubMed:9881970). Required for the generation of LTi (lymphoid tissue inducer) cells (PubMed:14691482). Regulates thymocyte survival through DNA-binding on ROREs of target gene promoter regions and recruitment of coactivaros via the AF-2 (PubMed:16148126). Also plays a key role, downstream of IL6 and TGFB and synergistically with RORA, for lineage specification of uncommitted CD4(+) T-helper (T(H)) cells into T(H)17 cells, antagonizing the T(H)1 program (PubMed:16990136, PubMed:18164222, PubMed:26607793). Probably regulates IL17 and IL17F expression on T(H) by binding to the essential enhancer conserved non-coding sequence 2 (CNS2) in the IL17-IL17F locus (PubMed:16990136, PubMed:18164222, PubMed:26607793). Plays a key role in tolerance to gut microbiota and foreign antigens in food by mediating differentiation of peripherally-induced regulatory T-cells (pTreg), which suppress inflammatory responses to commensal microorganisms and food proteins (PubMed:36070798, PubMed:36071167, PubMed:36071169, PubMed:40185101, PubMed:40298935). Specifically expressed in antigen-presenting cell populations, such as (1) type 3 innate lymphoid cells (ILC3s), which are required for tolerance to gut microbiota, (2) Thetis cells, also named Janus cells, which are enriched in gut lymph nodes during early life and are required for oral tolerance, and (3) a subset of dendritic cells (PubMed:36070798, PubMed:36071167, PubMed:36071169, PubMed:40185101, PubMed:40298935). Following activation by an unknown ligand in antigen-presenting cells, promotes recruitment of transcription regulators, such as PRDM16 or ZBTB46, and drives differentiation of pTreg cells, which maintain immune tolerance and prevent autoimmune diseases (PubMed:35831503, PubMed:40228524). May also play a role in the pre-TCR activation cascade leading to the maturation of alpha/beta T-cells and may participate in the regulation of DNA accessibility in the TCR-J(alpha) locus (PubMed:10602018).

Storage

Shipped at conditions	Blue Ice

Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C
Storage information	Do Not Freeze

Notes

ab232516 is the carrier-free version of ab207082.

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

What are the advantages of a recombinant monoclonal antibody?
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 batch production

For more information, read more on recombinant antibodies.

Conjugation ready
Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. 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 conjugation kits 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.

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

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

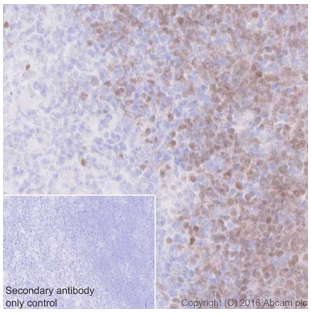
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

7 product images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ROR gamma antibody [EPR20006] - BSA and Azide free (ab232516)

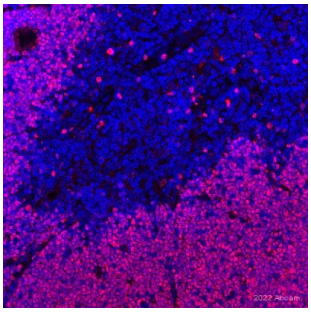
ROR gamma Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-ROR gamma antibody

Immunohistochemical analysis of paraffin-embedded mouse thymus tissue labeling ROR gamma with ab207082 at 1/3000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Nuclear staining on the cortical region of mouse thymus shown at a high magnification (400×) (PMID:14691482).
Counter stained 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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab207082).

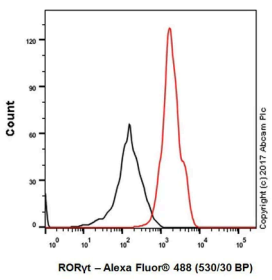
Heat mediated antigen retrieval was performed with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



This image is courtesy of an anonymous customer review

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ROR gamma antibody [EPR20006] - BSA and Azide free (ab232516)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of paraformaldehyde-fixed mouse thymus tissue staining with ab232516 at 5µg/ml dilution. Secondary antibody was Alexa Fluor® 555 Anti-Rabbit IgG at 2 µg/ml. Samples were incubated with the primary antibody with TBS + 0.1% Tween, 3% BSA, 5% horse serum for 16 hours at 4°C. Blocking was done using 3% BSA, 5% horse serum for 1 hour at 20 degrees. Heat mediated antigen retrieval with 10mM Tris Base, 1mM EDTA, pH 9.2. Hematoxylin was used for counterstaining.

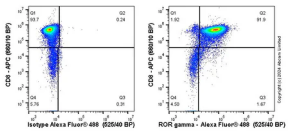


Flow Cytometry (Intracellular) - Anti-ROR gamma antibody [EPR20006] - BSA and Azide free (ab232516)

ROR gamma Flow Cytometry (Intracellular) staining using rabbit Anti-ROR gamma antibody

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed Mouse thymocytes labeling ROR gamma with ab207082 at 1/50 dilution. Goat anti rabbit IgG (Alexa Fluor® 488) at 1/2000 dilution was used as the secondary antibody.
Note: Cell surface staining with PE conjugated anti-mouse CD8 and Alexa Fluor® 647 conjugated anti mouse CD4 antibodies, followed by fixation with 4% PFA for 10 min and intracellular staining with ab207082. The image shows thymocytes in the CD4+CD8+ (red) or the CD4+CD8- (black, negative control) gate.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab207082).



Flow Cytometry (Intracellular) - Anti-ROR gamma antibody [EPR20006] - BSA and Azide free (ab232516)

ROR gamma Flow Cytometry (Intracellular) staining of Mouse thymocytes using rabbit Anti-ROR gamma antibody

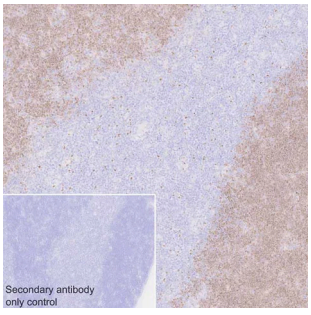
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab207082).

Flow cytometry overlay histogram showing mouse thymocytes stained with ab207082 (right) and isotype control (left).

The cells were incubated in 1x PBS containing 10µg/ml anti CD16/CD32 and 10% normal goat serum to block FC receptors and non-specific protein-protein interactions. The cells were then fixed and permeabilised with CytoFix/CytoPerm, followed by addition of the antibody (ab207082) (1x 10^6 in 100µl at 1.0µg/ml (1/2,000)) for 30min at 22°C.

The secondary antibody Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed was incubated at 1/2000 for 30min at 22°C

Acquisition of >30,000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter. Events were gated on live cells.



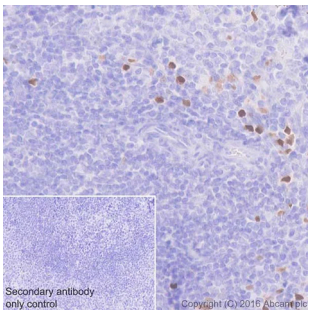
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ROR gamma antibody [EPR20006] - BSA and Azide free (ab232516)

ROR gamma Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-ROR gamma antibody

Immunohistochemical analysis of paraffin-embedded mouse thymus tissue labeling ROR gamma with ab207082 at 1/3000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Nuclear staining on mouse thymus cortex, located in the left and right of the image, with sporadic nuclear staining on medulla, middle region (PMID:14691482). Counter stained 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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab207082).

Heat mediated antigen retrieval was performed with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ROR gamma antibody [EPR20006] - BSA and Azide free (ab232516)

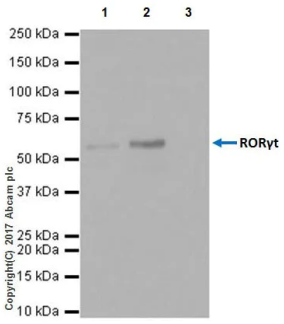
ROR gamma Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-ROR gamma antibody

Immunohistochemical analysis of paraffin-embedded mouse spleen tissue labeling ROR gamma with ab207082 at 1/3000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Sporadic nuclear staining on mouse spleen (PMID:14691482).
Counter stained 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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab207082).

Heat mediated antigen retrieval was performed with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunoprecipitation - Anti-ROR gamma antibody [EPR20006] - BSA and Azide free (ab232516)

ROR gamma was immunoprecipitated from 0.35 mg of Mouse thymus lysate with ab207082 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab207082 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/10000 dilution.
Lane 1: Mouse thymus lysate, 10 µg (Input).

Lane 2: ab207082 IP in Mouse thymus lysate.

Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab207082 inMouse thymus lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 10 seconds.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab207082).

All lanes:
Immunoprecipitation - Anti-ROR gamma antibody [EPR20006] (ab207082)

Predicted band size: 58 kDa

Observed band size: 58 kDa

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.