abcam

ab317453

Anti-VAChT antibody [EPR29154-71] - BSA and Azide free

Recombinant | RabMAb | Advanced Validation

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	EPR29154-71
Purification technique	Affinity purification Protein A
Concentration	1.015 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

mIHC

Tested

Species	Rat, Mouse
Dilution info	-

Notes	-

Not recommended

Species	Human
Dilution info	-
Notes	-

IHC-P

Tested

Species	Mouse, Rat
Dilution info	-
Notes	-

Not recommended

Species	Human
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Mouse
Dilution info	-
Notes	-

Expected

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

Not recommended

Species	Human
Dilution info	-
Notes	-

WB

Tested

Species	Mouse, Rat
Dilution info	-
Notes	-

Not recommended

Species	Human
Dilution info	-
Notes	-

IHC-Fr

Tested

Species	Mouse, Rat
Dilution info	-
Notes	-

Not recommended

Species	Human
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Mouse
Dilution info	-
Notes	-

Expected

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

Not recommended

Species	Human
Dilution info	-
Notes	-

IP

Tested

Species	Mouse
Dilution info	-
Notes	-

Expected

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

Not recommended

Species	Human
Dilution info	-
Notes	-

Target data

See full target information Slc18a3

Function	Electrogenic antiporter that exchanges one cholinergic neurotransmitter, acetylcholine or choline, with two intravesicular protons across the membrane of synaptic vesicles. Uses the electrochemical proton gradient established by the V-type proton-pump ATPase to store neurotransmitters inside the vesicles prior to their release via exocytosis (By similarity). Determines cholinergic vesicular quantal size at presynaptic nerve terminals in developing neuro-muscular junctions with an impact on motor neuron differentiation and innervation pattern (By similarity) (PubMed:19635813). Part of forebrain cholinergic system, regulates hippocampal synapse transmissions that underlie spatial memory formation (PubMed:23045697). Can transport serotonin.

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C

Notes

ab317453 is the carrirer-free version of ab317452.

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.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	The vesicular acetylcholine transporter often referred to as VAChT is responsible for packaging acetylcholine into vesicles in cholinergic neurons. This process is important for synaptic transmission in the nervous system. VAChT functions by using a proton gradient to transport acetylcholine against its concentration gradient into synaptic vesicles. VAChT has a molecular mass of approximately 55 kDa. It is widely expressed in cholinergic neurons throughout the central and peripheral nervous systems particularly in regions such as the cortex hippocampus and neuromuscular junctions.
Biological function summary	The function of VAChT plays an important role in the cholinergic system by facilitating neurotransmitter storage essential for neural communication. As part of the vesicular neurotransmitter transporter family VAChT collectively works with other proteins within this complex system to regulate acetylcholine availability and release. The stored acetylcholine within vesicles is released into the synaptic cleft in response to neuronal signals which promotes synapse firing and communication between neurons.
Pathways	Acetylcholine packaging and release managed by VAChT is integral to the cholinergic signaling pathway. This pathway is fundamental to ensuring proper function of learning memory and muscle control processes. VAChT works closely with the protein choline acetyltransferase (ChAT) which synthesizes acetylcholine from choline and acetyl-CoA. These proteins ensure a seamless conversion and transport loop within cholinergic neurons to maintain efficient neural signaling.
Associated diseases and disorders	VAChT has been linked to conditions that involve impaired cholinergic function such as Alzheimer's disease and myasthenia gravis. In Alzheimer's disease disruptions in the cholinergic system often marked by reduced VAChT expression contribute to cognitive decline. VAChT variations can also impact acetylcholine availability in myasthenia gravis a disorder characterized by muscle weakness. In both cases the relationship between VAChT and ChAT is important as ChAT levels often change in parallel affecting overall cholinergic signaling and response.

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.

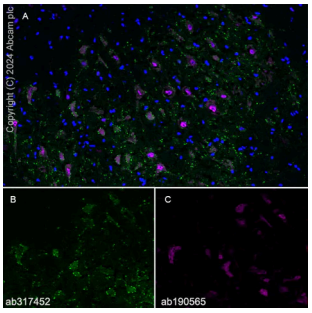
We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

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.

---

19 product images



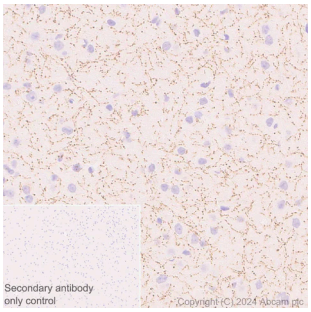
Immunohistochemistry (Frozen sections) - Anti-VAChT antibody [EPR29154-71] - BSA and Azide free (ab317453)

This data was developed using ab317452, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen Mouse cerebrum (fresh frozen) tissue labeling VAChT with ab317452 at 1/100 (5.08 ug/ml) dilution followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 (2 ug/mL) dilution (Green).

Panel A: merged staining of anti-VAChT (ab317452, green), anti-NeuN (ab190565, magenta) on mouse cerebrum.
Panel B: anti-VAChT stained on mouse cerebrum.
Panel C: anti-NeuN stained in neurons of mouse cerebrum.
The section was incubated in two rounds of staining: in the order of ab317452 and ab190565 for 1 hr at room temperature. The nuclear counterstain was DAPI (Blue). The section was then mounted using Fluoromount®.The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

Secondary antibody control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbedat 1/1000 (2 ug/mL) dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-VAChT antibody [EPR29154-71] - BSA and Azide free (ab317453)

This data was developed using ab317452, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Mouse cerebrum tissue labeling VAChT with ab317452 at 1/1000 (0.508 ug/ml) dilution, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

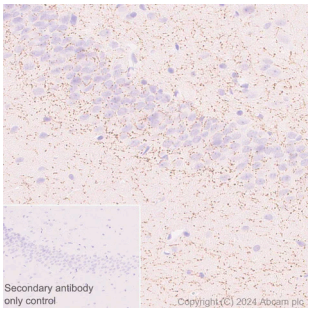
Positive staining on mouse cerebrum. The section was incubated with ab317452 for 30 mins at room temperature.
The immunostaining was performed on a Leica Biosystems BOND® RX instrument

Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins

---



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-VAChT antibody [EPR29154-71] - BSA and Azide free (ab317453)

This data was developed using ab317452, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Rat cerebrum tissue labeling VAChT with ab317452 at 1/1000 (0.508 ug/ml) dilution, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

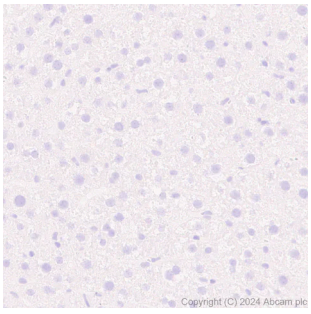
Positive staining on rat cerebrum. The section was incubated with ab317452 for 30 mins at room temperature.
The immunostaining was performed on a Leica Biosystems BOND® RX instrument

Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins

---



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-VAChT antibody [EPR29154-71] - BSA and Azide free (ab317453)

VAChT Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Rat liver using rabbit Anti-VAChT antibody

This data was developed using ab317452, the same antibody clone in a different buffer formulation.

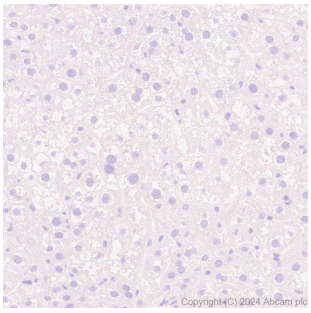
Immunohistochemical analysis of paraffin-embedded Rat liver tissue labeling VAChT with ab317452 at 1/1000 (0.508 ug/ml) dilution, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Low expression:: no staining on rat liver. The section was incubated with ab317452 for 30 mins at room temperature.
The immunostaining was performed on a Leica Biosystems BOND® RX instrument

Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-VAChT antibody [EPR29154-71] - BSA and Azide free (ab317453)

VAChT Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Mouse liver using rabbit Anti-VAChT antibody

This data was developed using ab317452, the same antibody clone in a different buffer formulation.

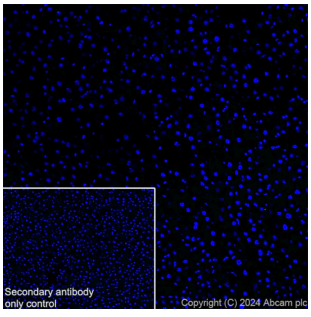
Immunohistochemical analysis of paraffin-embedded Mouse liver tissue labeling VAChT with ab317452 at 1/1000 (0.508 ug/ml) dilution, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Low expression:: no staining on mouse liver. The section was incubated with ab317452 for 30 mins at room temperature.
The immunostaining was performed on a Leica Biosystems BOND® RX instrument

Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins



Immunohistochemistry (Frozen sections) - Anti-VAChT antibody [EPR29154-71] - BSA and Azide free (ab317453)

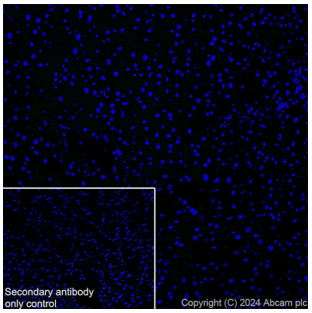
VAChT Immunohistochemistry (Frozen sections) staining of Rat liver (fresh frozen) using rabbit Anti-VAChT antibody

This data was developed using ab317452, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen Rat liver (fresh frozen) tissue labeling VAChT with ab317452 at 1/100 (5.08 ug/ml) dilution followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 (2 ug/mL) dilution (Green).

Low expression: confocal image showing no staining on rat liver (PMID: 9427309). The nuclear counterstain was DAPI (Blue). The section was incubated with ab317452 for 60 mins at room temperature. The section was then mounted using Fluoromount®.The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

Secondary antibody control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbedat 1/1000 (2 ug/mL) dilution.



Immunohistochemistry (Frozen sections) - Anti-VAChT antibody [EPR29154-71] - BSA and Azide free (ab317453)

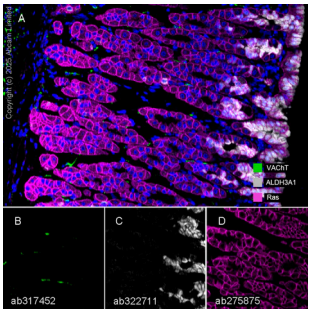
VAChT Immunohistochemistry (Frozen sections) staining of Mouse liver (fresh frozen) using rabbit Anti-VAChT antibody

This data was developed using ab317452, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen Mouse liver (fresh frozen) tissue labeling VAChT with ab317452 at 1/100 (5.08 ug/ml) dilution followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 (2 ug/mL) dilution (Green).

Low expression: confocal image showing no staining on mouse liver (PMID: 9427309). The nuclear counterstain was DAPI (Blue). The section was incubated with ab317452 for 60 mins at room temperature. The section was then mounted using Fluoromount®.The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

Secondary antibody control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbedat 1/1000 (2 ug/mL) dilution.



Multiplex immunohistochemistry - Anti-VAChT antibody [EPR29154-71] - BSA and Azide free (ab317453)

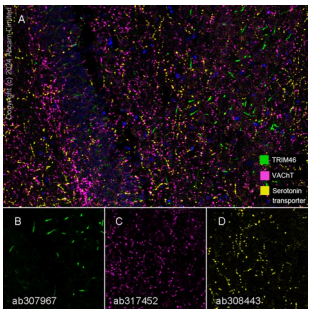
This data was developed using ab317452, the same antibody clone in a different buffer formulation.
Multiplex immunohistochemistry analysis of formalin/PFA-fixed paraffin-embedded mouse stomach tissue staining VAChT with ab317452 at 1/1000 dilution, ab322711 anti-ALDH3A1 used at 1/500 dilution and ab275875 anti-Ras used at 1/2000 dilution. Opal Polymer HRP Ms + Rb was used as a secondary antibody.

Panel A: merged staining of anti-VAChT (green; Opal™520), anti-ALDH3A1 (gray; Opal™570) and anti-Ras (magenta; Opal™690) on mouse stomach.
Panel B: anti-VAChT staining nerve plexus in mouse stomach.
Panel C: ant-ALDH3A1 staining epithelium in mouse stomach.
Panel D: ant-Ras showed mainly membranous staining in mouse stomach.
Nuclear DNA was labeled with DAPI (shown in blue).

The section was incubated in three rounds of staining: in the order of ab317452, ab322711 and ab275875 for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

The immunostaining was performed on a Leica Biosystems BOND®RX instrument with an Opal™ 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.



Multiplex immunohistochemistry - Anti-VAChT antibody [EPR29154-71] - BSA and Azide free (ab317453)

VAChT Multiplex immunohistochemistry staining of Rat hippocampus tissue using rabbit Anti-VAChT antibody

This data was developed using ab317452, the same antibody clone in a different buffer formulation.

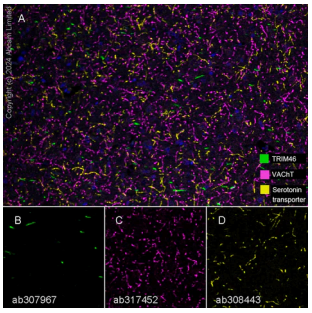
Multiplex immunohistochemistry analysis of formalin/PFA-fixed paraffin-embedded Rat hippocampus tissue staining TRIM46 with ab307967 at a 1:5000 (0.102 ug/ml) dilution, VAChT with ab317452 at 1:1000 (0.508 ug/ml) dilution and Serotonin transporter with ab308443 at a 1:500 (0.052 ug/ml) dilution followed by secondary antibody Opal Polymer HRP Ms + Rb.

Panel A: merged staining of anti-TRIM46 (green; Opal™520), anti-VAChT (magenta; Opal™690) and anti-Serotonin transporter (yellow; Opal™570) on rat hippocampus.Panel B: anti-TRIM46 staining the proximal part of the axon in rat hippocampus.Panel C: anti-VAChT staining vesicle in rat hippocampus.Panel D: anti-Serotonin transporter staining dendrites in rat hippocampus. Nuclear DNA was labeled with DAPI (shown in blue).

The section was incubated in three rounds of staining: in the order of ab307967, ab317452 and ab308443 for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal™ 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



Multiplex immunohistochemistry - Anti-VAChT antibody [EPR29154-71] - BSA and Azide free (ab317453)

VAChT Multiplex immunohistochemistry staining of Rat cerebrum tissue using rabbit Anti-VAChT antibody

This data was developed using ab317452, the same antibody clone in a different buffer formulation.

Multiplex immunohistochemistry analysis of formalin/PFA-fixed paraffin-embedded Rat cerebrum tissue staining TRIM46 with ab307967 at a 1:5000 (0.102 ug/ml) dilution, VAChT with ab317452 at 1:1000 (0.0.508 ug/ml) dilution and Serotonin transporter with ab308443 at a 1:500 (0.052 ug/ml) dilution followed by secondary antibody Opal Polymer HRP Ms + Rb.

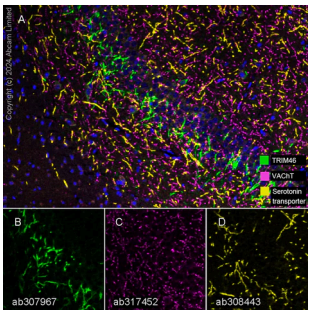
Panel A: merged staining of anti-TRIM46 (green; Opal™520), anti-VAChT (magenta; Opal™690) and anti-Serotonin transporter (yellow; Opal™570) on rat cerebrum.Panel B: anti-TRIM46 staining the proximal part of the axon in rat cerebrum.Panel C: anti-VAChT staining vesicle in rat cerebrum.Panel D: anti-Serotonin transporter staining dendrites in rat cerebrum. Nuclear DNA was labeled with DAPI (shown in blue).

The section was incubated in three rounds of staining: in the order of ab307967, ab317452 and ab308443 for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal™ 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins

---



Multiplex immunohistochemistry - Anti-VAChT antibody [EPR29154-71] - BSA and Azide free (ab317453)

VAChT Multiplex immunohistochemistry staining of Mouse hippocampus tissue using rabbit Anti-VAChT antibody

This data was developed using ab317452, the same antibody clone in a different buffer formulation.

Multiplex immunohistochemistry analysis of formalin/PFA-fixed paraffin-embedded Mouse hippocampus tissue staining TRIM46 with ab307967 at a 1:5000 (0.102 ug/ml) dilution, VAChT with ab317452 at 1:1000 (0.508 ug/ml) dilution and Serotonin transporter with ab308443 at a 1:500 (0.052 ug/ml) dilution followed by secondary antibody Opal Polymer HRP Ms + Rb.

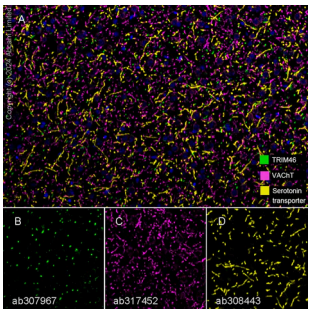
Panel A: merged staining of anti-TRIM46 (green; Opal™520), anti-VAChT (magenta; Opal™690) and anti-Serotonin transporter (yellow; Opal™570) on mouse hippocampus.Panel B: anti-TRIM46 staining the proximal part of the axon in mouse hippocampus.Panel C: anti-VAChT staining vesicle in mouse hippocampus.Panel D: anti-Serotonin transporter staining dendrites in mouse hippocampus.
Nuclear DNA was labeled with DAPI (shown in blue).

The section was incubated in three rounds of staining: in the order of ab307967, ab317452 and ab308443 for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal™ 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins

---



Multiplex immunohistochemistry - Anti-VAChT antibody [EPR29154-71] - BSA and Azide free (ab317453)

VAChT Multiplex immunohistochemistry staining of Mouse cerebrum tissue using rabbit Anti-VAChT antibody

This data was developed using ab317452, the same antibody clone in a different buffer formulation.

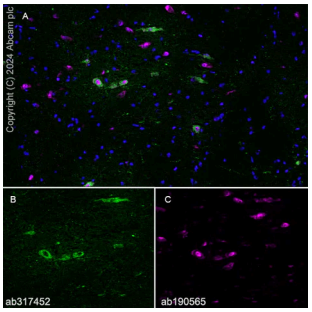
Multiplex immunohistochemistry analysis of formalin/PFA-fixed paraffin-embedded Mouse cerebrum tissue staining TRIM46 with ab307967 at a 1:5000 (0.102 ug/ml) dilution, VAChT with ab317452 at 1:1000 (0.508 ug/ml) dilution and Serotonin transporter with ab308443 at a 1:500 (0.052 ug/ml) dilution followed by secondary antibody Opal Polymer HRP Ms + Rb.

Panel A: merged staining of anti-TRIM46 (green; Opal™520), anti-VAChT (magenta; Opal™690) and anti-Serotonin transporter (yellow; Opal™570) on mouse cerebrum.Panel B: anti-TRIM46 staining the proximal part of the axon in mouse cerebrum.Panel C: anti-VAChT staining vesicle in mouse cerebrum.Panel D: anti-Serotonin transporter staining dendrites in mouse cerebrum.
Nuclear DNA was labeled with DAPI (shown in blue).

The section was incubated in three rounds of staining: in the order of ab307967, ab317452 and ab308443 for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal™ 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



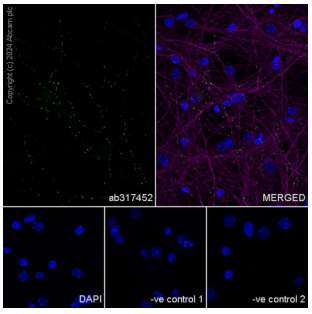
Immunohistochemistry (Frozen sections) - Anti-VAChT antibody [EPR29154-71] - BSA and Azide free (ab317453)

This data was developed using ab317452, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen Rat cerebrum (fresh frozen) tissue labeling VAChT with ab317452 at 1/100 (5.08 ug/ml) dilution followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 (2 ug/mL) dilution (Green).

Panel A: merged staining of anti-VAChT (ab317452, green), anti-NeuN (ab190565, magenta) on rat cerebrum.
Panel B: anti-VAChT stained on rat cerebrum.
Panel C: anti-NeuN stained in neurons of rat cerebrum.
The section was incubated in two rounds of staining: in the order of ab317452 and ab190565 for 1 hr at room temperature. The nuclear counterstain was DAPI (Blue). The section was then mounted using Fluoromount®.The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

Secondary antibody control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbedat 1/1000 (2 ug/mL) dilution.



Immunocytochemistry/ Immunofluorescence - Anti-VAChT antibody [EPR29154-71] - BSA and Azide free (ab317453)

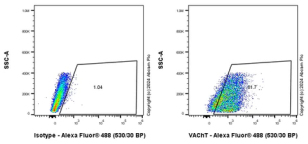
This data was developed using ab317452, the same antibody clone in a different buffer formulation.

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized mouse hippocampal neuron cells labelling VAChT with ab317452 at 1/100 (5.08 ug/ml) dilution, followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 (2 ug/ml) dilution (Green).

Confocal image showing cytoplasmic staining in mouse hippocampal neuron (shown in green). The counterstain was observed in magenta. Nuclear DNA was labeled with DAPI (shown in blue). Confocal scanning Z step was set as 0.3 µm followed by image processing with maximum Z projection.
Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

ab11267 Anti-MAP2 mouse monoclonal antibody was used to counterstain at 1/500 (4ug/ml) dilution (Magenta). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 (2 ug/ml) dilution.

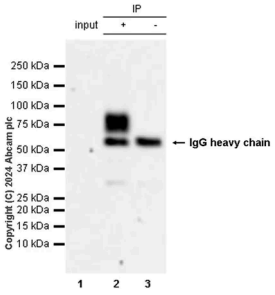


Flow Cytometry (Intracellular) - Anti-VAChT antibody [EPR29154-71] - BSA and Azide free (ab317453)

This data was developed using ab317452, the same antibody clone in a different buffer formulation.

Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized mouse primary neuron cells labelling VAChT with ab317452 at 1/500 dilution (0.1 ug)/Right compared with a Rabbit monoclonal IgG (ab172730) / Left isotype control.

Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/5000 dilution was used as the secondary antibody.



Immunoprecipitation - Anti-VAChT antibody [EPR29154-71] - BSA and Azide free (ab317453)

This data was developed using ab317452, the same antibody clone in a different buffer formulation.

VAChT was immunoprecipitated from 0.35 mg Mouse brain tissue lysate with ab317452 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab317452 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP)(ab131366) was used at 1/5000 dilution.

Lane 1: Mouse brain tissue lysate
Lane 2: ab317452 IP in Mouse brain tissue lysate
Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab317452 in mouse brain whole cell lysate

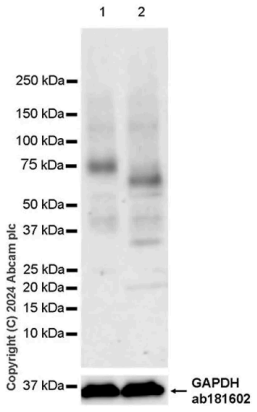
All lanes:
Immunoprecipitation - Anti-VAChT antibody [EPR29154-71] (ab317452) at 1/30 dilution

All lanes:
Mouse brain tissue lysate

Secondary

All lanes:
Immunoprecipitation - VeriBlot for IP Detection Reagent (HRP) (ab131366) at 1/5000 dilution

Exposure time: 58s



Western blot - Anti-VAChT antibody [EPR29154-71] - BSA and Azide free (ab317453)

This data was developed using ab317452, the same antibody clone in a different buffer formulation.
VAChT is a glycoprotein of approximately 75 kDa and detected as a 60-kDa band after treated with Peptide:N-glycosidase F (PNGase F).

In Western blot, Anti-GAPDH antibody [EPR16891] - Loading Control (ab181602) staining at 1/200000 dilution.

This blot was developed using a high sensitivity ECL substrate. The high-sensitivity ECL substrate used allows for the detection of proteins in the mid-femtogram range.

All lanes:
Western blot - Anti-VAChT antibody [EPR29154-71] (ab317452) at 1/1000 dilution

Lane 1:
Untreated Mouse brain tissue lysate at 60 µg

Lane 2:
Mouse brain tissue lysate treated with Peptide:N-glycosidase F (PNGase F) at 60 µg

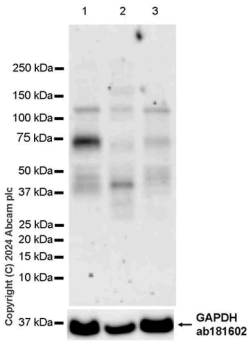
Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Developed using the ECL technique.

Observed band size: 60 kDa, 75 kDa, 36 kDa

Exposure time: 180s



Western blot - Anti-VAChT antibody [EPR29154-71] - BSA and Azide free (ab317453)

VAChT Western blot staining using rabbit Anti-VAChT antibody

This data was developed using ab317452, the same antibody clone in a different buffer formulation.

Low expression: liver, cerebellum (PMID: 9427309).

The expression profile/ molecular weight observed is consistent with what has been described in the literature (PMID: 22087075, PMID: 7559575, PMID: 34230437).

The identity of the bands higher than 100 kDa and lower than 50 kDa are unknown.

In Western blot, Anti-GAPDH antibody [EPR16891] - Loading Control (ab181602) staining at 1/200000 dilution.

All lanes:
Western blot - Anti-VAChT antibody [EPR29154-71] (ab317452) at 1/1000 dilution

Lane 1:
Rat brain tissue lysate at 60 µg

Lane 2:
Rat liver tissue lysate at 60 µg

Lane 3:
Rat cerebellum tissue lysate at 60 µg

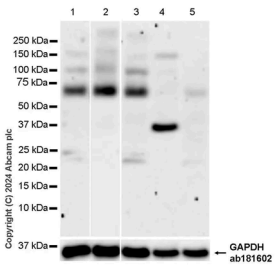
Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Developed using the ECL technique.

Observed band size: 75 kDa, 36 kDa

Exposure time: 180s



Western blot - Anti-VAChT antibody [EPR29154-71] - BSA and Azide free (ab317453)

VAChT Western blot staining using rabbit Anti-VAChT antibody

This data was developed using ab317452, the same antibody clone in a different buffer formulation.

Low expression: liver, kidney (PMID: 9427309).

The expression profile/ molecular weight observed is consistent with what has been described in the literature (PMID: 22087075, PMID: 7559575, PMID: 34230437).

The identity of the bands higher than 100 kDa (in lanes 1-4) are unknown.

The identity of the lower MW band at approximately 37 kDa (in lane 4) is unknown.

In Western blot, Anti-GAPDH antibody [EPR16891] - Loading Control (ab181602) staining at 1/200000 dilution.

Exposure time: Lane 2: 26 seconds, lanes 1, 3-5: 114 seconds

All lanes:
Western blot - Anti-VAChT antibody [EPR29154-71] (ab317452) at 1/1000 dilution

Lane 1:
Mouse brain tissue lysate at 80 µg

Lane 2:
Mouse retina tissue lysate at 80 µg

Lane 3:
Mouse hippocampus tissue lysate at 80 µg

Lane 4:
Mouse liver tissue lysate at 80 µg

Lane 5:
Mouse kidney tissue lysate at 80 µg

Secondary

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

Observed band size: 75 kDa, 36 kDa

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