

IBA1 Polyclonal Antibody

Catalog NumberPA5-27436

Product data sheet

Details		Species Reactivity	
Size	100 µL	Species reactivity	Human, Mouse, Rat
Host/Isotope	Rabbit / IgG	Published species	Dog, Rat, Cat, Mouse, Human, Not Applicable
Class	Polyclonal	Tested Applications	
Type	Antibody	Dilution *	
Immunogen	Recombinant protein encompassing a sequence within the center region of human Iba1	Flow Cytometry (Flow)	1:50-1:200
Conjugate	Unconjugated	Immunohistochemistry (Paraffin) (IHC (P))	1:100-1:1,000
Form	Liquid	Immunoprecipitation (IP)	1:100-1:500
Concentration	0.14 mg/mL	Western Blot (WB)	1:500-1:10,000
Purification	Antigen affinity chromatography	Immunocytochemistry (ICC/IF)	1:100-1:1,000
Storage buffer	PBS, pH 7, with 1% BSA, 20% glycerol	Published Applications	
Contains	0.025% ProClin 300	Western Blot (WB)	See 4 publications below
Storage Conditions	Store at 4°C short term. For long term storage, store at -20°C, avoiding freeze/thaw cycles.	Immunohistochemistry (IHC)	See 9 publications below
		Immunocytochemistry (ICC/IF)	See 1 publications below
		Immunohistochemistry (Paraffin) (IHC (P))	See 1 publications below
		Miscellaneous PubMed (Misc)	See 1 publications below
		Immunohistochemistry - Free Floating (IHC (Free))	See 1 publications below

* Suggested working dilutions are given as a guide only. It is recommended that the user titrate the product for use in their own experiment using appropriate negative and positive controls.

Product specific information

Predicted reactivity: Bovine, Pig, Rhesus Monkey (>80% identity)

Background/Target Information

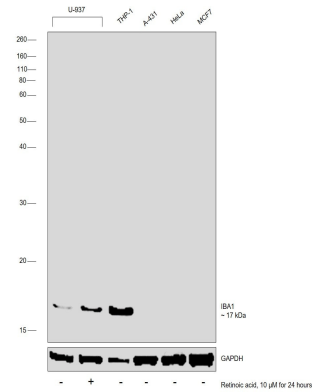
AIF1 is induced by cytokines and interferon. It is thought to be involved in the negative regulation of growth of vascular smooth muscle cells, which contributes to the anti-inflammatory response to vessel wall trauma.

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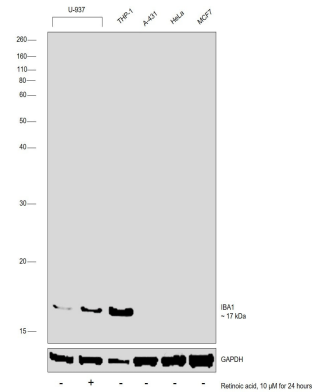
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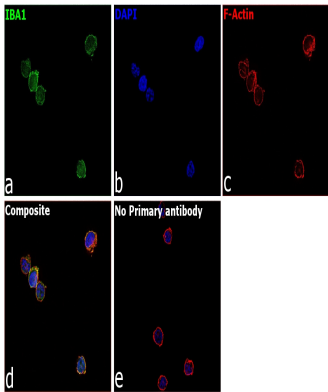
IBA1 Antibody (PA5-27436) in WB

Western blot was performed using Anti-IBA1 Polyclonal Antibody (Product # PA5-27436) and a 17 kDa band corresponding to IBA1 was observed across U-937, U-937 treated with Retinoic Acid and THP-1. Whole Cell Extract-WCL (30 µg lysate) of U-937 (Lane 1), U-937 treated with Retinoic Acid (10 µM for 24 hours) (Lane 2), THP-1 (Lane 3), A-431 (Lane 4), HeLa (Lane 5) and MCF7 (Lane 6), were electrophoresed using NuPAGE™ 12% Bis-Tris Protein Gel (Product # NP0342BOX). Resolved proteins were then transferred onto a Nitrocellulose membrane (Product # IB23002) by iBlot® 2 Dry Blotting System (Product # IB21001). The blot was probed with the primary antibody (1:2000 dilution) and detected by chemiluminescence with Goat anti-Rabbit IgG (Heavy Chain) Superclonal™ Recombinant Secondary Antibody, HRP (Product # A27036,1:4000 dilution) using the iBright FL 1000 (Product # A32752). Chemiluminescent detection was performed using Novex® ECL Chemiluminescent Substrate Reagent Kit (Product # WP20005).



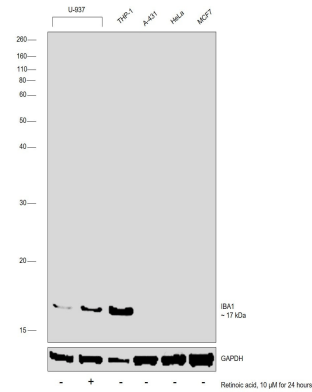
IBA1 Antibody (PA5-27436)

Antibody specificity was demonstrated by detection of differential basal expression of the target across cell lines tested owing to their inherent genetic constitution. Relative expression of IBA1 was observed in THP-1 in comparison to A-431, HeLa and MCF7 using Anti-IBA1 Polyclonal Antibody (Product # PA5-27436) in Western Blot. {RE}



IBA1 Antibody (PA5-27436) in ICC/IF

Immunofluorescence analysis of IBA1 was performed using 70% confluent log phase THP-1 cells. The cells were fixed with 4% paraformaldehyde for 15 minutes, permeabilized with 0.1% Triton™ X-100 for 10 minutes, and blocked with 2% BSA for 1 hour at room temperature. The cells were labeled with IBA1 Polyclonal Antibody (Product # PA5-27436) at 1:500 dilution in 0.1% BSA, incubated at 4 degree celsius overnight and then labeled with Donkey anti-Rabbit IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor Plus 488 (Product # A32790, 1:2000 dilution) for 45 minutes at room temperature (Panel a: Green). Nuclei (Panel b:Blue) were stained with ProLong™ Diamond Antifade Mountant with DAPI (Product # P36962). F-actin (Panel c: Red) was stained with Rhodamine Phalloidin (Product # R415, 1:300 dilution). Panel d represents the merged image showing membrane and cytoplasmic localization. Panel e represents control cells with no primary antibody to assess background. The images were captured at 60X magnification.



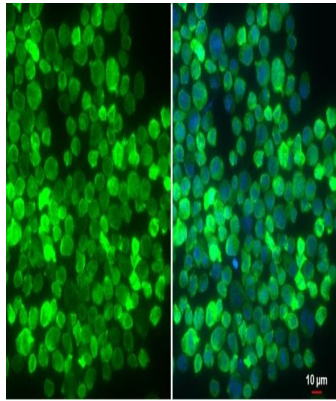
IBA1 Antibody (PA5-27436)

Altered expression of proteins upon cell treatment demonstrates antibody specificity. Western blot using IBA1 Polyclonal Antibody (Product #PA5-27436), shows increased expression of IBA1 in U-937 cell line upon Retinoic acid treatment. {TM}

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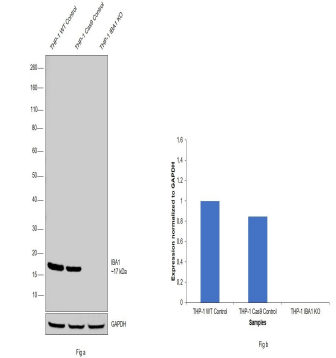
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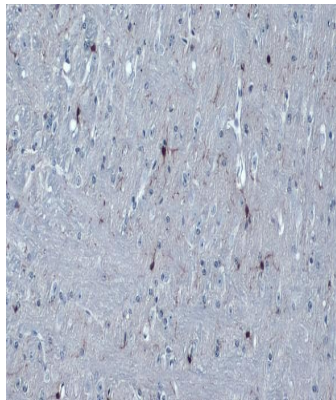
IBA1 Antibody (PA5-27436) in ICC/IF

IBA1 Polyclonal Antibody detects Iba1 protein at cell membrane by immunofluorescent analysis. Sample: THP-1 cells were fixed in 4% paraformaldehyde at RT for 15 min. Green: Iba1 stained by IBA1 Polyclonal Antibody (Product # PA5-27436) diluted at 1:500. Red: alpha Tubulin, a cytoskeleton marker, stained by alpha Tubulin antibody (Product # PA5-85158) diluted at 1:1,000. Blue: Fluoroshield with DAPI .



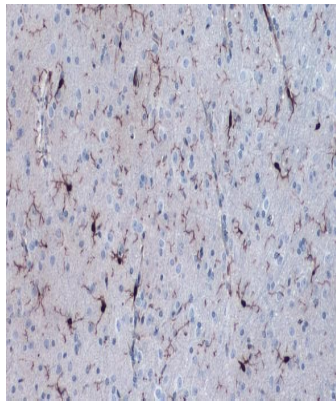
IBA1 Antibody (PA5-27436)

Antibody specificity was demonstrated by CRISPR-Cas9 mediated knockout of target protein. A loss of signal was observed for target protein in IBA1 KO cell line compared to control cell line using Anti-IBA1 Polyclonal Antibody (Product # PA5-27436). {KO}



IBA1 Antibody (PA5-27436) in IHC (P)

IBA1 Polyclonal Antibody detects Iba1 protein at cell membrane and cytoplasm by immunohistochemical analysis. Sample: Paraffin-embedded mouse cerebellum. Iba1 stained by IBA1 Polyclonal Antibody (Product # PA5-27436) diluted at 1:1,000. Antigen Retrieval: Citrate buffer, pH 6.0, 15 min.



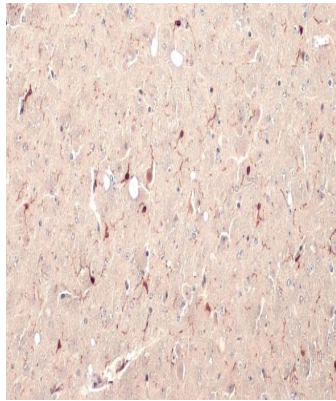
IBA1 Antibody (PA5-27436) in IHC (P)

IBA1 Polyclonal Antibody detects Iba1 protein at cell membrane and cytoplasm by immunohistochemical analysis. Sample: Paraffin-embedded rat cerebellum. Iba1 stained by IBA1 Polyclonal Antibody (Product # PA5-27436) diluted at 1:1,000. Antigen Retrieval: Citrate buffer, pH 6.0, 15 min.

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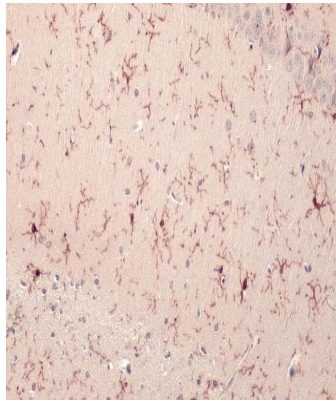
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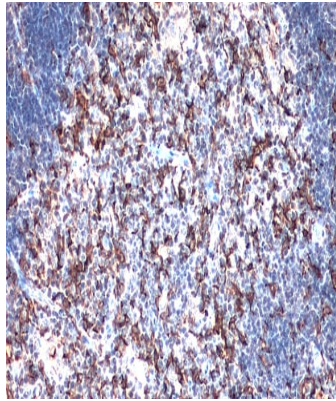
IBA1 Antibody (PA5-27436) in IHC (P)

IBA1 Polyclonal Antibody detects Iba1 protein at cell membrane and cytoplasm by immunohistochemical analysis. Sample: Paraffin-embedded rat brain. Iba1 stained by IBA1 Polyclonal Antibody (Product # PA5-27436) diluted at 1:500. Antigen Retrieval: Citrate buffer, pH 6.0, 15 min.



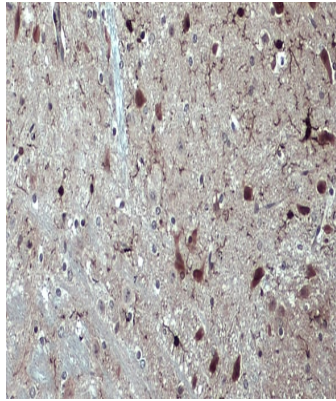
IBA1 Antibody (PA5-27436) in IHC (P)

IBA1 Polyclonal Antibody detects Iba1 protein at cell membrane and cytoplasm by immunohistochemical analysis. Sample: Paraffin-embedded mouse brain. Iba1 stained by IBA1 Polyclonal Antibody (Product # PA5-27436) diluted at 1:500. Antigen Retrieval: Citrate buffer, pH 6.0, 15 min.



IBA1 Antibody (PA5-27436) in IHC (P)

Immunohistochemistry (Paraffin) analysis of IBA1 was performed in paraffin-embedded mouse thymus gland tissue using IBA1 Polyclonal Antibody (Product # PA5-27436) at a dilution of 1:500. Antigen Retrieval: Citrate buffer, pH 6.0, 15 min.



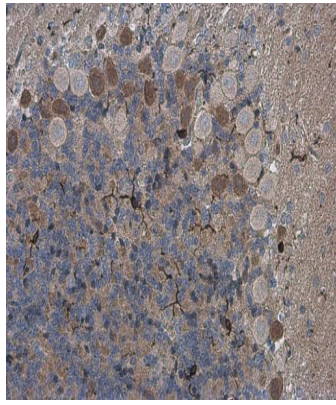
IBA1 Antibody (PA5-27436) in IHC (P)

Immunohistochemistry (Paraffin) analysis of IBA1 was performed in paraffin-embedded mouse brain tissue using IBA1 Polyclonal Antibody (Product # PA5-27436) at a dilution of 1:500. Antigen Retrieval: Citrate buffer, pH 6.0, 15 min.

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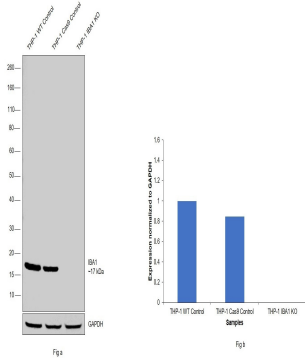


IBA1 Antibody (PA5-27436) in IHC (P)

Immunohistochemistry (Paraffin) analysis of IBA1 was performed in paraffin-embedded rat brain tissue using IBA1 Polyclonal Antibody (Product # PA5-27436) at a dilution of 1:500.

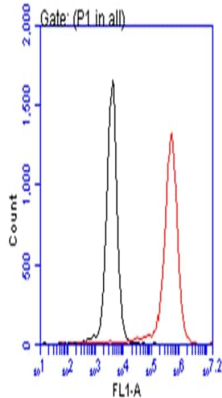
IBA1 Antibody (PA5-27436) in WB

Knockout of IBA1 was achieved by CRISPR-Cas9 genome editing using LentiArray™ Lentiviral sgRNA (Product # A32042, AssayID CRISPR741364_LV) and LentiArray Cas9 Lentivirus (Product # A32064). Western blot analysis of IBA1 was performed by loading 30 µg of THP-1 wild type (Lane 1), THP-1 CAS9 (Lane 2), THP-1 IBA1 KO (Lane 3) whole cell extracts. The blot was probed with Anti-IBA1 Polyclonal Antibody (Product # PA5-27436) using 1:2000 dilution and Goat anti-Rabbit IgG (Heavy Chain), Superclonal™ Recombinant Secondary Antibody, HRP (Product # A27036). Loss of signal upon CRISPR mediated knockout (KO) using the LentiArray™ CRISPR product line confirms that antibody is specific to IBA1.



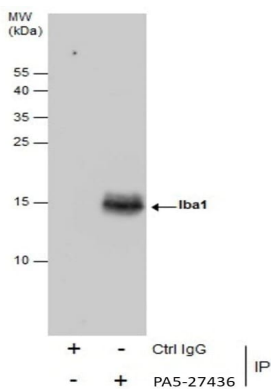
IBA1 Antibody (PA5-27436) in Flow

Flow Cytometry analysis of NQO1 was performed in THP-1 cells using NQO1 Monoclonal Antibody (A180) (Product # MA1-91897) (red) at a dilution of 1:50. Black: Unlabelled sample was used as a control. Acquisition of 20,000 events were collected using a Dylight 488-conjugated secondary antibody for FACS analysis.



IBA1 Antibody (PA5-27436) in IP

Immunoprecipitation of Iba1 was performed in K562 whole cell extracts using 5 µg of IBA1 Polyclonal Antibody (Product # PA5-27436). Samples were transferred to a membrane and probed with IBA1 Polyclonal Antibody as a primary antibody and an HRP-conjugated anti-Rabbit IgG was used as a secondary antibody.



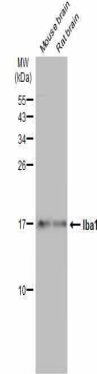
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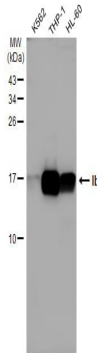
IBA1 Antibody (PA5-27436) in WB

Western blot analysis of IBA1 was performed by separating 50 µg of various tissue extracts by 15% SDS-PAGE. Proteins were transferred to a membrane and probed with a IBA1 Polyclonal Antibody (Product # PA5-27436) at a dilution of 1:1000. The HRP-conjugated anti-rabbit IgG antibody was used to detect the primary antibody.



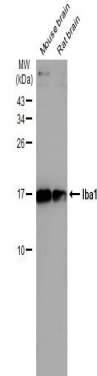
IBA1 Antibody (PA5-27436) in WB

Western Blot using IBA1 Polyclonal Antibody (Product # PA5-27436). Various whole cell extracts (30 µg) were separated by 15% SDS-PAGE, and the membrane was blotted with IBA1 Polyclonal Antibody (Product # PA5-27436) diluted at 1:500. The HRP-conjugated anti-rabbit IgG antibody was used to detect the primary antibody.



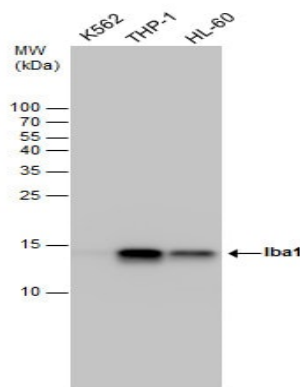
IBA1 Antibody (PA5-27436) in WB

Western Blot using IBA1 Polyclonal Antibody (Product # PA5-27436). Various tissue extracts (50 µg) were separated by 15% SDS-PAGE, and the membrane was blotted with IBA1 Polyclonal Antibody (Product # PA5-27436) diluted at 1:500. The HRP-conjugated anti-rabbit IgG antibody was used to detect the primary antibody.



IBA1 Antibody (PA5-27436) in WB

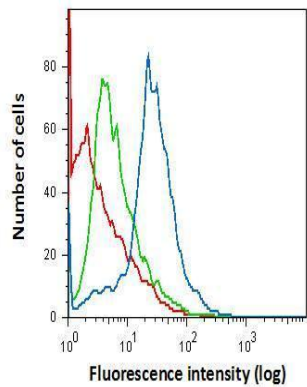
Western Blot analysis of IBA1 was performed by separating 30 µg of various whole cell extracts by 15% SDS-PAGE. Proteins were transferred to a membrane and probed with a IBA1 Polyclonal Antibody (Product # PA5-27436) at a dilution of 1:5000 and a HRP-conjugated anti-rabbit IgG secondary antibody.



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IBA1 Antibody (PA5-27436) in Flow

Flow cytometry on primary murine microglia cells, staining with IBA1 Polyclonal Antibody (Product # PA5-27436) using 1.0 µg per 4x10⁵ cells. (Product # PA5-27436) (blue), Rabbit IgG (green) ,Unstained (red).

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PubMed References For IBA1 Polyclonal Antibody

4 Western Blot References

Species / Dilution	Summary
Rat / 1:500	PA5-27436 was used in Western Blot to examine markers for inflammation and brain plasticity in the hippocampi of low-capacity runner rats and compared them to the more physically fit high-capacity runner rats.
	Brain, behavior, and immunity (2021; 97: 250) "Rats bred for low intrinsic aerobic exercise capacity link obesity with brain inflammation and reduced structural plasticity of the hippocampus." Author(s):Mäkinen E,Lensu S,Honkanen M,Laitinen P,Wikgren J,Koch LG,Britton SL,Kainulainen H,Pekkala S,Nokia MS PubMed Article URL: http://dx.doi.org/10.1016/j.bbi.2021.06.017
Rat / 1:1000	PA5-27436 was used in Immunohistochemistry (Paraffin), Western Blot to evaluate the potential benefit of neural progenitor cells for recovering nerve injury.
	International journal of molecular sciences (2021; 22:) "Human Pluripotent Stem Cell-Derived Neural Progenitor Cells Promote Retinal Ganglion Cell Survival and Axon Recovery in an Optic Nerve Compression Animal Model." Author(s):Park M,Kim HM,Shin HA,Lee SH,Hwang DY,Lew H PubMed Article URL: http://dx.doi.org/10.3390/ijms222212529
Mouse / 1:1000	PA5-27436 was used in Western Blotting to indicate that PFF-induced pathology could lead to astrocyte and/or microglia senescence in PD brains, which may contribute to neuropathology in this model.
	Cells (2021; 10:) "Alpha-Synuclein Preformed Fibrils Induce Cellular Senescence in Parkinson's Disease Models." Author(s):Verma DK,Seo BA,Ghosh A,Ma SX,Hernandez-Quijada K,Andersen JK,Ko HS,Kim YH PubMed Article URL: http://dx.doi.org/10.3390/cells10071694
Mouse / 1:1000	PA5-27436 was used in Western Blotting to evaluate the participation of spinal cord glial cells in the pathophysiology of pain induced by Leishmania amazonensis infection in BALB/c mice.
	Journal of neuroinflammation (2019; 16:) "Contribution of spinal cord glial cells to L. amazonensis experimental infection-induced pain in BALB/c mice." Author(s):Borghi SM,Fattori V,Pinho-Ribeiro FA,Domiciano TP,Miranda-Sapla MM,Zaninelli TH,Casagrande R,Pinge-Filho P,Pavanelli WR,Alves-Filho JC,Cunha FQ,Cunha TM,Verri WA PubMed Article URL: http://dx.doi.org/10.1186/s12974-019-1496-2

9 Immunohistochemistry References

Species / Dilution	Summary
Mouse / 1:1000	PA5-27436 was used in Immunohistochemistry to suggest that this AgRP neural circuit plays a unique role in persistent control of energy expenditure and body weight, hinting next-generation therapeutic approaches for obesity and metabolic disorders.
	Nature communications (2021; 12:) "Deciphering an AgRP-serotonergic neural circuit in distinct control of energy metabolism from feeding." Author(s):Han Y,Xia G,Srisai D,Meng F,He Y,Ran Y,He Y,Farias M,Hoang G,Tóth I,Dietrich MO,Chen MH,Xu Y,Wu Q PubMed Article URL: http://dx.doi.org/10.1038/s41467-021-23846-x
Mouse / 1:200	PA5-27436 was used in Immunohistochemistry to determine whether dexmedetomidine protects against SAE and whether 2 adrenoceptor plays a role in this protection.
	Brain, behavior, and immunity (2021; 91: 296) "Dexmedetomidine attenuates sepsis-associated inflammation and encephalopathy via central 2A adrenoceptor." Author(s):Mei B,Li J,Zuo Z PubMed Article URL: http://dx.doi.org/10.1016/j.bbi.2020.10.008
Mouse / Not Cited	PA5-27436 was used in Immunohistochemistry to support TDO inhibition as a potential therapeutic strategy to decrease motor, cognitive, and gastrointestinal symptoms in Parkinson's disease.
	The FEBS journal (2021; 288: 4311) "Pharmacological validation of TDO as a target for Parkinson's disease." Author(s):Perez-Pardo P,Grobben Y,Willemsen-Seegers N,Hartog M,Tutone M,Muller M,Adolfs Y,Pasterkamp RJ,Vu-Pham D,van Doornmalen AM,van Cauter F,de Wit J,Gerard Sterrenburg J,Uitdehaag JCM,de Man J,Buijsman RC,Zaman GJR,Kraneveld AD PubMed Article URL: http://dx.doi.org/10.1111/febs.15721

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	<p>PA5-27436 was used in Immunohistochemistry-immunofluorescence to investigate the expression of type 1 interferon pathway members in CDV-induced cerebellar lesions to gain more information into their role in lesion development.</p>
Dog / 1:500	<p>International journal of molecular sciences (2019; 20:) "Interferon-Stimulated Genes-Mediators of the Innate Immune Response during Canine Distemper Virus Infection." Author(s):Klotz D,Gerhauser I PubMed Article URL:http://dx.doi.org/10.3390/ijms20071620</p>
Mouse / 1:500	<p>PA5-27436 was used in Immunohistochemistry to introduce astrocytes as an important component of pain gating by activation of A-fibers, which thus exert nonneuronal control of pain.</p> <p>Science advances (2021; 7:) "Astrocytes contribute to pain gating in the spinal cord." Author(s):Xu Q,Ford NC,He S,Huang Q,Anderson M,Chen Z,Yang F,Crawford LK,Caterina MJ,Guan Y,Dong X PubMed Article URL:http://dx.doi.org/10.1126/sciadv.abi6287</p>
Mouse / Not Cited	<p>PA5-27436 was used in Immunohistochemistry to suggest that the activation of the NOX2 pathway, possibly mediated by macrophage infiltration, plays a pivotal role in progressive renal fibrosis in pcy mice.</p> <p>Nephron (2022; 146: 393) "Upregulation of NADPH Oxidase 2 Contributes to Renal Fibrosis in pcy Mice: An Experimental Model of Nephronophthisis." Author(s):Hoshino Y,Sonoda H,Mikoda N,Ikeda M PubMed Article URL:http://dx.doi.org/10.1159/000520697</p>
Mouse / 1:1000	<p>PA5-27436 was used in Immunohistochemistry to study the pathogenic mechanisms of GM1-gangliosidosis in a novel -galactosidase (knockout mouse model.</p> <p>Journal of clinical medicine (2020; 9:) "Axonopathy and Reduction of Membrane Resistance: Key Features in a New Murine Model of Human G<sub>M1</sub>-Gangliosidosis." Author(s):Eikelberg D,Lehmbecker A,Brogden G,Tongtako W,Hahn K,Habierski A,Hennemann JB,Naim HY,Felmy F, Baumgärtner W,Gerhauser I PubMed Article URL:http://dx.doi.org/10.3390/jcm9041004</p>
Mouse / Not Cited	<p>PA5-27436 was used in Immunohistochemistry to show that NADPH oxidase 2-induced oxidative stress accompanied by macrophage infiltration plays a role in alteration of oxygen homeostasis in Aqp11-/- mice.</p> <p>Biochemistry and biophysics reports (2019; 17: 169) "Involvement of the NADPH oxidase 2 pathway in renal oxidative stress in <i>Aqp11</i> <sup>-/-</sup> mice." Author(s):Hoshino Y,Sonoda H,Nishimura R,Mori K,Ishibashi K,Ikeda M PubMed Article URL:http://dx.doi.org/10.1016/j.bbrep.2019.01.003</p>
Not Applicable / 1:500	<p>PA5-27436 was used in Immunohistochemistry to report a hereditary leukodystrophy in Standard Schnauzer puppies caused by TSEN54:c.371G>A.</p> <p>PLoS genetics (2019; 15:) "TSEN54 missense variant in Standard Schnauzers with leukodystrophy." Author(s):Störk T,Nessler J,Anderegg L,Hünerfauth E,Schmutz I,Jagannathan V,Kyöstilä K,Lohi H,Baumgärtner W,Tipold A,Leeb T PubMed Article URL:http://dx.doi.org/10.1371/journal.pgen.1008411</p>

1 Immunocytochemistry References	
Species / Dilution	Summary
	<p>PA5-27436 was used in Immunocytochemistry-immunofluorescence to provide insights into the involvement of epigenetics in intellectual disability.</p>
Mouse / Not Cited	<p>Development (Cambridge, England) (2021; 148:) "The histone demethylase PHF8 regulates astrocyte differentiation and function." Author(s):Iacobucci S,Padilla N,Gabrielli M,Navarro C,Lombardi M,Vicioso-Mantis M,Verderio C,de la Cruz X,Martínez-Balbás MA PubMed Article URL:http://dx.doi.org/10.1242/dev.194951</p>
1 Immunohistochemistry (Paraffin) References	
Species / Dilution	Summary
	<p>PA5-27436 was used in Immunohistochemistry (Paraffin) to lead to the diagnosis of primary diffuse leptomeningeal oligodendrogliomatosis.</p>
Cat / 1:1000	<p>Frontiers in veterinary science (2022; 8:) "Case Report: Primary Diffuse Leptomeningeal Oligodendrogliomatosis in a Young Adult Cat." Author(s):Chludzinski E,Puff C,Weber J,Hewicker-Trautwein M PubMed Article URL:http://dx.doi.org/10.3389/fvets.2021.795126</p>
1 Miscellaneous PubMed References	

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Species / Dilution	Summary
Rat / 1:2400	PA5-27436 was used in western blot to investigate mechanisms associated with chronic pain after SCI
	Experimental neurology (2016; 278: 91) "Analysis of the behavioral, cellular and molecular characteristics of pain in severe rodent spinal cord injury." Author(s):Lee-Kubli CA,Ingves M,Henry KW,Shiao R,Collyer E,Tuszynski MH,Campana WM PubMed Article URL: http://dx.doi.org/10.1016/j.expneurol.2016.01.009
1 Immunohistochemistry - Free Floating References	
Species / Dilution	Summary
Mouse / Not Cited	PA5-27436 was used in immunohistochemistry - free floating to investigate the contribution of TRPM2 to beta-amyloid neuronal toxicity
	The Journal of neuroscience : the official journal of the Society for Neuroscience (2015; 35: 15157) "The Transient Receptor Potential Melastatin 2 (TRPM2) Channel Contributes to -Amyloid Oligomer-Related Neurotoxicity and Memory Impairment." Author(s):Ostapchenko VG,Chen M,Guzman MS,Xie YF,Lavine N,Fan J,Beraldo FH,Martyn AC,Belrose JC,Mori Y,MacDonald JF,Prado VF,Prado MA,Jackson MF PubMed Article URL: http://dx.doi.org/10.1523/JNEUROSCI.4081-14.2015