Mouse monoclonal antibody to rat capsaicin receptor (VR1, TRPV1, 819-838), [Clone BS397]: IgG

Catalogue No.:	M-1714-100
Description:	The capsaicin receptor (VR1, TRPV1) is a ligand-activated non-selective calcium permeant cation channel involved in detection of noxious chemical and thermal stimuli. The receptor seems to mediate proton influx and may be involved in intracellular acidosis in nociceptive neurons. It is involved in mediation of inflammatory pain and hyperalgesia. Sensitized by a phosphatidylinositol second messenger system activated by receptor tyrosine kinases, which involves PKC isozymes and PCL. Activation by vanilloids, like capsaicin, and temperatures higher than 42 degrees Celsius, exhibits a time- and Ca2+-dependent outward rectification, followed by a long-lasting refractory state. Mild extracellular acidic pH (6.5) potentiates channel activation by noxious heat and vanilloids, whereas acidic conditions (pH less than 6) directly activate the channel. Can be activated by endogenous compounds, including 12-hydroperoxytetraenoic acid and bradykinin. Acts as ionotropic endocannabinoid receptor with central neuromodulatory effects. Triggers a form of long-term depression (TRPV1-LTD) mediated by the endocannabinoid anandamine in the hippocampus and nucleus accumbens by affecting AMPA receptors endocytosis (Ref: uniprot.org).
Unit size:	100 ug
Antigen:	A synthetic peptide (C-GSLKPEDAEVFKDSMVPGEK) as a part of the C-terminal rat VR1 protein (aa: 819-838) has been used as the immunogen.
Sequence:	C-GSLKPEDAEVFKDSMVPGEK; aa 819-838 rat VR1
Antigen Location:	C-terminal
Antigen Length:	20 amino acids
Antibody Type:	Mouse monoclonal IgG
Isotype:	IgG2b, k-light chain
Clone:	BS397
Other Names:	VR1; Transient receptor potential cation channel subfamily V member 1; TrpV1; osm-9-like TRP channel 1; OTRPC1; Vanilloid receptor 1; Capsaicin receptor; VR-1
Accession:	Uniprot: O35433; TRPV1_RAT
Produced in:	Mouse
Molecular Weight:	Monomer 90-100kDa in mouse brain extracts; dimer 180-200kDa can be observed under some conditions
Purity:	Protein G purified mouse immunoglobulin
Applications:	Flow Cytometry: 2 ug/10^6 cells. Western blotting: 0.5-2 ug/mL , SDS-PAGE on Bis-Tris gel 4-12%, 5% beta-mercaptoethanol, primary antibody O/N incubation in 5% skim milk/TBST. Secondary is anti-mouse-HRP, 1/6000 dilution, 2h at room temperature. Blot developed on Li-Cor? C-DiGit? lot Scanner. IHC: Frozen or PEG embedded tissues tested (PEG embedding, see Klosen P et al (1993) J Histochem Cytochem. 41(3):455-63). Conditions tested: 1-10 ug/mL in PBS, 48 hours, followed by detection via directly conjugated fluorescent anti-mouse

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	secondary. Antibody not yet tested on paraffin embedded sections. Other immunohistochemistry methods not yet tested but are expected to be reactive. ICC: 4% formaldehyde fixed cells tested; requires permeabilization step as antigen epitope is intracellular. Suggested primary antibody concentration: 1-2 ug/mL.Biosensis recommends optimal dilutions/concentrations should be determined by the end user.
Specificity:	Antibody is specific for rat/mouse VR1 protein in westerns and immunofluorescent immunohistochemistry on mouse PEG fixed DRG tissues. Pre-absorption with immunogen obliterates positive staining. Cross reactivity with other non-VR1 proteins is minimal; cross reactivity with VR1 from other species not yet tested.
Species Against:	Rat/mouse, other species not yet tested
Antibody Against:	Rat VR1
Cross-reactivity:	This antibody clone is known to react with rat and mouse TrpV1. It is predicted to react with guinea pig due to sequence homology.
Form:	Lyophilized from PBS, pH 7.4 with 3% trehalose.
Appearance:	Dry powder
Reconstitution:	Reconstitute in 100 uL of sterile water. Centrifuge to remove any insoluble material. Final buffer contains no preservatives but will contain 3% trehalose and buffer salts.
Storage:	Store lyophilized antibody at 2-8C. After reconstitution divide in to aliquots and store at -20C for a higher stability. Antibody contains no preservatives. Storage at 2-8C with an appropriate antibacterial agent. USE Sterile methods. Highest purity Glycerol (1:1) may be added for an additional stability when stored at refrigerated or freezing temperatures. Avoid repetitive freeze/thaw cycles.
Expiry Date:	12 months after purchase if unopened.
Specific References:	Bai J et al. (2018) [EXPRESS] Attenuation of TRPV1 by AMG-517 after Nerve Injury Promotes Peripheral Axonal Regeneration in Rats. Mol Pain. 2018 Jan 1; [Epub ahead of print]. Application: WB
General References:	Peng H.Y. (2008) TRPV1 mediates the uterine capsaicin-induced NMDA NR2B-dependent cross-organ reflex sensitization in anesthetized rats. Am J Physiol Renal Physiol. Nov;295(5):F1324-35.

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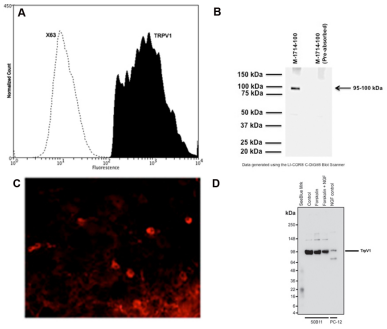

A: Analysis of TRPV1 expression in rat PC12 cell line by Flow Cytometry. Fixing and permeabilization of cells: Absolute methanol (10 minutes in ice) and 0.1% Tween-20 in PBS, Blocking: 1% BSA, Primary antibody: Mouse Monoclonal antibody to TRPV1 (cat # M-1714-100, 2 µg per ~10^6 cells) for 30 minutes at RT, Secondary antibody: Goat anti-mouse PE labeled secondary antibody (1:100 dilution), 20 minutes in dark at room temperature. Negative control: Non-specific Control IgG, clone X63 (cat # M-1249-200, black dashed). Data and results were generated using Orflo MoxiflowTM instrument and protocols.

B: Western blot of TrpV1 in rat PC12 cell lysates (80 µg/lane). M-1714-100 detects TrpV1 protein at 95-100 kDa. SDS-PAGE: denatured and reduced; Transfer: Tris-Glycine buffer; Membrane: nitrocellulose (0.45 µm); Blocking: 5% skim milk in TBST, 1 hour at RT; Primary antibody: overnight at 4°C (2 µg/mL); Secondary antibody: anti-mouse-HRP (1/6000) 2 hours at RT; Detection: Chemiluminiscence.

C: Immunohistochemical staining of TrpV1 in mouse dorsal root ganglia. Immunoreactivity was visualized with anti-mouse-Cy3 conjugate (red). Magnification: 20x. Courtesy P. Vilimas, Flinders University Adelaide.

D: Western blot (denatured and reduced) of TrpV1 in cell lysates of forskolin and NGF stimulated 50B11 hybrid mouse x rat DRG cell lines and NGF-stimulated PC12 cells (10 µg/lane). M-1714-100 detects monomeric TrpV1 protein at 95-100 kDa. Primary antibody: 1 µg/mL (4°C overnight). Detection: Chemiluminiscence. Courtesy Dr. D. Matusica, Flinders University.

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