ADVANCED TARGETING SYSTEMS

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Anti-SERT-SAP

TARGETED SAP CONJUGATE

[antibody to serotonin re-uptake transporter (SERT)]-saporin
targets serotonin re-uptake transporter (SERT)

Catalog Number: IT-23
Quantity: 25 micrograms, 100 micrograms, 250 micrograms
Format: PBS (0.14 M Sodium Chloride; 0.003 M Potassium Chloride; 0.002 M Potassium Phosphate; 0.01 M Sodium Phosphate; pH 7.4), no preservative. Sterile-filtered.
Host: Mouse

Background:
Targeted SAP conjugates are powerful and specific lesioning agents used in the technique known as Molecular Surgery. The ribosome-inactivating protein, saporin (from the seeds of the plant, *Saponaria officinalis*) is bound to a targeting agent (anything that is recognized on the cell surface and internalized). The targeted conjugate is administered to cells (*in vitro* or *in vivo*). The targeting agent seeks out and binds to its target on the cell surface. The conjugate is internalized, saporin breaks away from the targeting agent, and inactivates the ribosomes which causes protein inhibition and, ultimately, cell death. Cells that do not have the cell surface marker are not affected.
Anti-SERT-SAP utilizes a monoclonal antibody to the fourth extracellular domain of the serotonin re-uptake transporter (SERT). The sequence of the peptide antigen is identical in rat, mouse, human, and other mammalian species. SERT is the major determinant of serotonin inactivation following release at synapses, is the site of action for many tricyclic antidepressants and the SSRIs (serotonin-selective reuptake inhibitors), and is also targeted by a number of psychostimulants including cocaine, methylphenidate, and MDMA 'ecstasy.' SERT is produced from a single gene and is expressed in both the CNS and GI system.
Decreased serotonergic neurotransmission has been proposed to play a key role in the etiology of depression. Recent findings suggest that SERT might be linked to both neurotic and sexual behavior as well as to obsessive-compulsive disorder (OCD). Anti-SERT-SAP specifically eliminates cells expressing SERT making it an excellent tool for studying the sertonergic system which is known to modulate mood, emotion, sleep and appetite and thus is implicated in the control of numerous behavioral and physiological functions.

Specificity and Preparation:
This targeted toxin recognizes cells that express SERT. The sequence of the peptide antigen is identical in rat, mouse, human, and other mammalian species. Anti-SERT-SAP is a chemical conjugate of a monoclonal antibody to the fourth extracellular domain of serotonin re-uptake transporter (SERT) and the ribosome-inactivating protein, saporin. This product is routinely tested by cytotoxicity assay.

Usage and Storage:
Anti-SERT-SAP specifically eliminates SERT-expressing cells *in vitro* and *in vivo*. All other cells are left untouched. **There may be lot-to-lot variation in material; working dilutions must be determined by end user. If this is a new lot, you must assess the proper working dilution before beginning a full experimental protocol.**
Gently spin down material before use; 5-10 seconds in a microfuge should be adequate. Store the material in undiluted aliquots at –20°C for 1-2 months. For longer term storage store the material at –80°C. Material should be aliquoted to a convenient volume and quantity to avoid repeated freezing and thawing that can damage the protein content. Under these conditions, the material has a very stable shelf-life. Thawing should be done at room temperature or on ice. The thawed solution should remain on ice until use.
Do not use a reducing agent (such as dithiothreitol, beta-mercaptoethanol or ascorbic acid) with this material. It will inactivate the toxin.
This material is an extremely potent cytotoxin. Handling should be done by experienced personnel. Gloves and safety glasses are required when handling this product. Care in disposal is mandatory; autoclaving or exposure to 0.2 M sodium hydroxide will inactivate the material. All labware that comes into contact with this material

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should be likewise treated.

Available Control(s): Mouse IgG-SAP, Antibody to SERT

References:

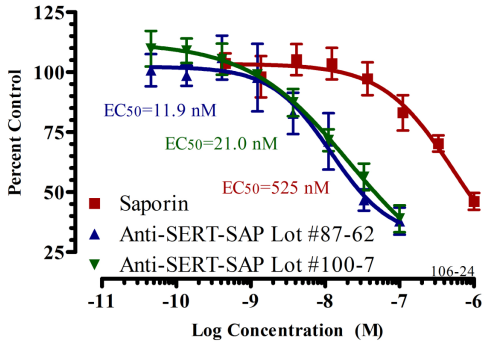
1. Nattie EE, Li A, Richerson G, Lappi D (2002) Specific killing of rat medullary raphe 5-HT neurons by a serotonin transporter antibody-saporin conjugate reduced the ventilatory response to increased CO2 during sleep and wakefulness. *Soc Neurosci Mtg*, Orlando FL, Abstract #221.3.
2. Lappi D, Kohls M, Majer K, Russell B, Blakely R, Richerson G (2002) Targeting serotonin re-uptake transporter (SERT)-expressing cells with a monoclonal antibody to an epitope from the extracellular domain of SERT: Results with a saporin conjugate. *4th Forum of European Neuroscience, Paris FRANCE*, Abstract #049.7.
3. Kohls MD, Majer KA, Russell BJ, Han Q, Blakely RD, Lappi DA (2001) A monoclonal antibody to an extracellular domain of the serotonin transporter: Characterization and targeting properties. *Soc Neurosci Mtg*, San Diego CA, Abstract #814.9.

Safety:

Good laboratory technique must be employed for safe handling of this product.
This requires observation of the following practices:

1. Wear appropriate laboratory attire, including lab coat, gloves and safety glasses.
2. Do not pipet by mouth, inhale, ingest or allow product to come into contact with open wounds. Wash thoroughly any part of the body which comes into contact with the product.
3. Avoid accidental autoinjection by exercising extreme care when handling in conjunction with any injection device.
4. This product is intended for research use by qualified personnel only. It is not intended for use in humans or as a diagnostic agent. Advanced Targeting Systems is not liable for any damages resulting from the misuse or handling of this product.

To view protocol(s) for this and other products please visit: www.ATSbio.com/protocols



RBL-2H3 cells cloned for sensitivity to anti-SERT-SAP were plated at 1000 cells/90 µl/well and incubated overnight. Anti-SERT-SAP and saporin were added in 10 µl volumes, and the plates incubated for 72 hours. The cells were fixed with 10% TCA, then stained with 0.4% sulfarhodamine B/1% acetic acid. The plates were read at 564 nm. Data analysis was done by Prism software (GraphPad, San Diego).