



Anti-DAT-SAP TARGETED SAP CONJUGATE

a tool for eliminating cells that express dopamine transporter (DAT-ECD) in rat or human; targeted via a monoclonal antibody to the second extracellular loop of DAT, eliminated via saporin

Catalog Number: IT-25

Quantity: 25 micrograms, 100 micrograms, 250 micrograms, 1 milligram

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: Rat

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.

The dopamine transporter (DAT) is a sodium-dependent reuptake carrier that is suspected to play a role in such neurologic and psychiatric disorders as Parkinson's disease, Tourette's syndrome, schizophrenia, and addiction. It is a 12-transmembrane domain transporter with the N- and C-terminus regions located within the cytoplasm. Because of its importance in dopamine use, DAT is an identifier of dopaminergic neurons.

Anti-DAT-SAP is highly specific for cells that express DAT, including mesencephalic dopamine neurons of the substantia nigra, pars compacta, and ventral tegmental area. Specific elimination of cells that express DAT is useful in studying the role of dopaminergic neurons in one of the devastating diseases of old age, Parkinson's.

Specificity and Preparation:

This targeted toxin recognizes cells that express DAT in rat and human. The toxin is a chemical conjugate of a rat monoclonal antibody to the second extracellular loop of the dopamine transporter (DAT-ECD, Cat. #AB-N17) and the ribosome-inactivating protein, saporin (Cat. #PR-01).

Usage and Storage:

Anti-DAT-SAP is highly specific for cells that express DAT, including mesencephalic dopamine neurons of the substantia nigra, pars compacta, and ventral tegmental area. There may be lot-to-lot variation in material; working dilutions must be determined by end user. If this is a new lot, you <u>must</u> 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 should be likewise treated.



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Available Control(s): Antibody to Dopamine Transporter (DAT-ECD), Rat IgG-SAP

References:

- 1. Wiley RG, Harrison MB, Levey A, Lappi DA (2003) Destruction of midbrain dopaminergic neurons by using an immunotoxin to the dopamine transporter. *Cell Mol Neurobiol* 23:839-850.
- 2. Lappi DA (2003) A new immunotoxin for targeting dopaminergic neurons. Targeting Trends 4(3):1.

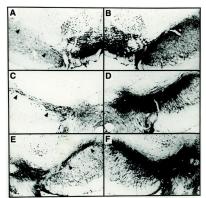
Safety:

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/support/protocols



Representative sections stained for tyrosine hydroxylase from rats with intrastriatal injections of anti-DAT-SAP 2 weeks prior to sacrifice. Panes A, C, and E are ipsilateral to the striatal injections. Panels B, D, and F are contralateral, from the same sections. The anti-DAT-SAP doses were 2.8 mg in A, 0.56 mg in C, and 0.28 mg in E. Note loss of dopaminergic neurons from the ipsilateral substantia nigra, pars compacta (arrowheads) with the greatest extent of cell loss in A and the least in E. The magnification bar in F indicates 100 mm and applies to all panels.