ADVANCED TARGETING SYSTEMS

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Anti-CD44-SAP

TARGETED SAP CONJUGATE

a tool for eliminating cells that express all isoforms of the CD44 receptor in multiple species; targeted via the rat monoclonal antibody to mouse CD44 (clone IM7), eliminated via saporin

Catalog Number: IT-72
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.
CD44 is a cell-surface glycoprotein that plays a role in cell-cell interactions, cell adhesion and migration. CD44 is a receptor for hyaluronic acid and also interacts with other ligands, such as osteopontin, collagens, and matrix metalloproteinases. CD44 participates in a wide variety of cellular functions such a lymphocyte activation, recirculation and homing, hematopoieses, and tumor metastasis. CD44 has been considered an activity marker and potential novel therapeutic target in multiple sclerosis and is associated with relapses in non-small cell lung cancers.

Specificity and Preparation:
This targeted toxin recognizes cells that expressing all isoforms of the CD44 receptor with expected reactivity in mouse, human, baboon, monkey, bovine, cat, dog, ferret, horse, pig, cynomolgus, cynomolgus monkey, and rhesus monkey. Anti-CD44-SAP is a bonded toxin between biotinylated rat monoclonal antibody to mouse CD44 (clone IM7) and the secondary conjugate Streptavidin-ZAP (IT-27) containing the ribosome-inactivating protein, saporin.

Usage and Storage:
Anti-CD44-SAP eliminates cells expressing all isoforms of CD44. 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. 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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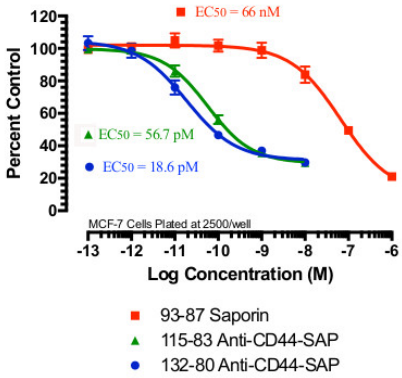
Available Control(s): Rat IgG-SAP

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



MCF-7 cells were plated at 2500 cells/90 ul/well and incubated overnight. Saporin (PR-01), and Anti-CD44-SAP dilutions were made in cell media and 10 ul was added to each well. The plates incubated 72 hours. To develop the plates, the medium was removed and 50 ul of 0.4% sulforhodamine-B in 1% acetic acid was added to each well and the plates incubated for 30 minutes at room temperature. The plates were then washed three times with 1% acetic acid and allowed to dry. The dye was solubilized with 100 ul of 10 mM Tris-Base per well, with 5 minutes of gentle shaking. The plates were read at 564 nm and data analysis was done with Prism software (GraphPad, San Diego).