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Rat IgG-SAP NON-TARGETED SAPORIN CONTROL MOLECULE

[pre-immune rat IgG antibody]-saporin

targets no known cells; used as a control with saporin conjugates

Catalog Number: IT-17

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: Rat

Background:

Controls are a vital part of the scientific procedure; without them it is difficult to isolate the specific effects from the non-specific or artifactual. This control molecule is the same molecular weight, consists of similar, comparable materials and is synthesized with the same protocols as the targeted toxins. The difference is the cell-specific targeting agents are replaced with "blanks," antibodies or peptides that have no specificity, and no ability to target cells. In short, they are the perfect control molecules for behavioral experiments with Advanced Targeting Systems' targeted toxins.

Specificity and Preparation:

This control conjugate has no known specificity; it may react with cells that possess Fc receptors. Rat IgG-SAP is a chemical conjugate of pre-immune rat IgG antibody and the ribosome inactivating protein, saporin. The product is routinely tested by cytotoxicity assay.

Usage and Storage:

Rat IgG-SAP serves as a control for rat IgG-containing immunolesioning agents (Mac-1-SAP, Anti-DAT-SAP, Anti-CD25-SAP mouse). 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. The material should be stored at -20°C in undiluted aliquots. 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. The material can be handled safely using normal laboratory precautions.

Do not use a reducing agent (such as dithiothreitol, beta-mercaptoethanol or ascorbic acid) with this material. It will inactivate the toxin.

For disposal: autoclave, or expose to 0.2 M NaOH, materials that come into contact with the toxin.



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References:

- 1. Tazzari PL, Bolognesi A, De Totero D, Falini B, Lemoli RM, Soria MR, Pileri S, Gobbi M, Stein H, Flenghi L, *et al.* (1992) Ber-H2 (anti-CD30)-saporin immunotoxin: a new tool for the treatment of Hodgkin's disease and CD30+ lymphoma: *in vitro* evaluation. *Brit J Haemat* 81:203-211.
- 2. Dinota A, Tazzari PL, Michieli M, Visani G, Gobbi M, Bontadini A, Tassi C, Fanin R, Damiani D, Grandi M, *et al.* (1990) *In vitro* bone marrow purging of multidrug-resistant cells with a mouse monoclonal antibody directed against Mr 170,000 glycoprotein and a saporin-conjugated anti-mouse antibody. *Cancer Res* 50:4291-4294.
- 3. Thorpe PE, Brown AN, Bremner JA Jr, Foxwell BM, Stirpe F (1985) An immunotoxin composed of monoclonal anti-Thy 1.1 antibody and a ribosome-inactivating protein from *Saponaria officinalis*: potent antitumor effects *in vitro* and *in vivo*. *J Natl Cancer Inst* 75(1):151-159.

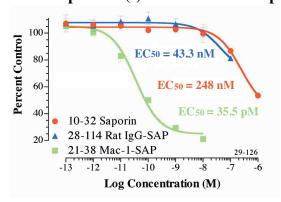
Safety:

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



WEHI-274.1 cells, a murine monocytic cell line, were plated at 2500 cells per well in 90 microliters of medium. After allowing acclimatization overnight, the cells were exposed to the various reagents at the indicated concentrations for 72 hours. MTS (Promega) was added and after two hours, plates were read at 492 nM on a Molecular Diagnostics Spectramax 340 plate reader with SoftMax software. Data were analyzed by Prism 3.0 software.