ADVANCED TARGETING SYSTEMS

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Fab IgG-SAP

NON-TARGETED SAPORIN CONTROL MOLECULE

a tool for use as control for goat IgG-containing immunolesioning agents; non-targeted via pre-immune goat IgG Fab conjugated to saporin

Catalog Number: IT-67
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: Goat

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 conjugates. 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 conjuates.
Fab IgG-SAP serves as a control for goat IgG-containing immunolesioning agents and specifically all Fab-ZAP secondary conjugates. The secondary Fab-ZAP antibody conjugates use a monovalent Fab with your primary antibody to determine if the primary antibody can internalize saporin and would, therefore, be suitable for conjugation as a primary immunotoxin. Fab IgG-SAP, used in place of the secondary antibody conjugate, will give a definitive baseline for comparison of the activity of the primary antibody to internalize.

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

Usage and Storage:
Fab IgG-SAP serves as a control for goat IgG-containing immunolesioning agents and specifically all Fab-ZAP secondary conjugates. **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. 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:

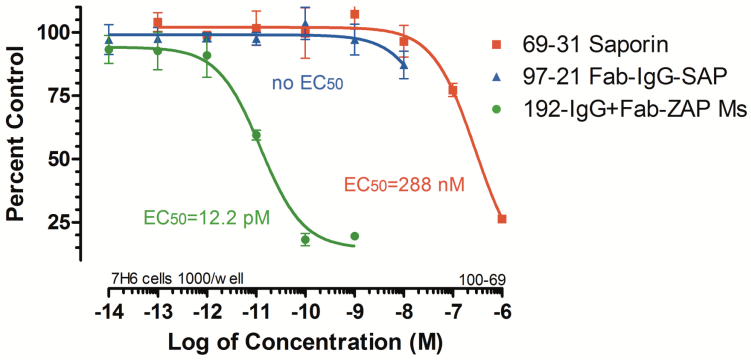
1. Tazzari PL, Bolognesi A, De Totero D, Falini B, Lemoli RM, Soria MR, Pileri S, Gobbi M, Stein H, Flenghi L, *et al*. (1992) Ber-H2 (anti-CD30)-saporin immunotoxin: a new tool for the treatment of Hodgkin's disease and CD30+ lymphoma: *in vitro* evaluation. *Brit J Haemat* 81:203-211.
2. Dinota A, 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/support/protocols



7H6 cells, a clone of the rat glial cell line C6, were plated at 1000 cells/90 µl/ well and incubated overnight. Saporin (PR -01) and Fab-IgG-SAP (IT-67) dilutions were made into cell media and 10 µl was added to each well. 192-IgG antibody (AB-N43) was diluted in cell media containing, at a final concentration, 45 ng/10 µl Fab-ZAP Mouse (IT-48) and 10 µl was added to each well. The plates incubated for 72 hours. The plates were then developed with a XTT/PMS solution and left to incubate for 2 hours at 37°C. Plates were read at 450 nm in a plate reader and data analysis was done with Prism software (GraphPad, San Diego).