

Anti-Dinitrophenyl-KLH Antibodies

- A-6430** anti-dinitrophenyl-KLH, rabbit IgG fraction
A-11097 anti-dinitrophenyl-KLH, rabbit IgG fraction, Alexa Fluor® 488 conjugate
A-6435 anti-dinitrophenyl-KLH, rabbit IgG fraction, biotin-XX conjugate
A-6423 anti-dinitrophenyl-KLH, rabbit IgG fraction, fluorescein conjugate

Quick Facts

Storage upon receipt:

- 4°C or -20° (in aliquots)
- Avoid freeze-thaw cycles
- Protect fluorescent conjugates from light

Abs/Em of A-6423 and A-11097: 494/520 nm

Introduction

Because of their high affinity for the dinitrophenyl hapten^{1,2} (DNP), Molecular Probes' antibodies against the dinitrophenyl-keyhole limpet hemocyanin conjugate (anti-DNP-KLH) are sensitive probes for detecting DNP-labeled molecules.³⁻⁶ The fluorescent and biotinylated anti-DNP-KLH conjugates can serve as second-step reagents for signal amplification in protein and nucleic acid detection systems, including *in situ* hybridization studies employing DNP-modified DNA probes.

For labeling nucleic acids and proteins with DNP, Molecular Probes offers DNP-X succinimidyl ester (D-2248) and the Fluor-Reporter® Biotin/DNP Protein Labeling Kit (F-6348). For detecting biotinylated anti-DNP-KLH in enzyme-linked hybridization and immunoassays using avidin or streptavidin bridge techniques, we prepare a biotin conjugate of horseradish peroxidase (P-917).

For assays not requiring a fluorescent conjugate of the anti-DNP antibody, Molecular Probes offers unconjugated anti-DNP-KLH antibody (A-6430). This antibody can be used for Fc receptor-mediated delivery of DNP-labeled liposomes prepared from DNP phospholipids.⁷ Because anti-DNP antibody binds well to DAMP (D-1552), an acidotropic reagent that can penetrate cell membranes, anti-DNP-KLH antibodies can also be used with DAMP for immunochemical localization of intracellular compartments with low pH.⁸⁻¹¹ In addition, anti-DNP-KLH

antibodies may recognize compounds with a trinitrophenyl group, allowing the isolation of cell-surface molecules of human melanoma cells that have been labeled with trinitrobenzene sulfonic acid (TNBS).¹²

Contents and Storage

The anti-DNP-KLH antibodies are supplied in unit sizes of 0.5 mL as a 2 mg/mL solution. The conjugates (A-6423, A-6435 and A-11097) are in 0.1 M sodium phosphate, 0.1 M sodium chloride, pH 7.5, containing 5 mM sodium azide. The unlabeled antibody (A-6430) is in phosphate-buffered saline (PBS), pH 7.2, containing 5 mM sodium azide. For the conjugates, the degree of labeling is indicated on the product label. The anti-DNP-KLH antibodies are stable for two to three months when stored undiluted at 4°C. For longer storage, divide the solution into aliquots and freeze at -20°C. These aliquots should be stable for at least one year when stored frozen. PROTECT FLUORESCENT CONJUGATES FROM LIGHT. AVOID REPEATED FREEZING AND THAWING.

Properties

Molecular Probes' anti-DNP-KLH antibody was raised in rabbit against DNP conjugated to keyhole limpet hemocyanin. The IgG fraction, which contains both anti-DNP and anti-KLH IgGs, was isolated from total serum and determined to be >95% pure by SDS-polyacrylamide gel electrophoresis.

It is a good practice to centrifuge protein conjugate solutions briefly in a microcentrifuge before use; only the supernatant should then be added to the experiment. This step will eliminate any protein aggregates that may have formed during storage, thereby reducing nonspecific background staining.

Because staining protocols vary with application, the appropriate dilution of antibody should be determined empirically.¹³ Peak absorption and fluorescence emission wavelengths for both the fluorescein anti-DNP-KLH and Alexa Fluor® 488 anti-DNP-KLH conjugates are 494 nm and 520 nm, respectively.

References

1. J Exp Med 145, 931 (1977); 2. Adv Immunol 2, 1 (1962); 3. J Immunol Methods 150, 193 (1992); 4. Biotechniques 9, 186 (1990); 5. J Histochem Cytochem 38, 69 (1990); 6. Science 247, 64 (1990); 7. Proc Natl Acad Sci USA 77, 4089 (1980); 8. J Cell Biol 117, 1211 (1992); 9. Nature 352, 70 (1991); 10. J Histochem Cytochem 38, 1927 (1990); 11. J Cell Biol 107, 2137 (1988); 12. Biotechniques 9, 324 (1990); 13. Harlow, E. and Lane, D., *Antibodies: A Laboratory Manual*, Cold Spring Harbor Laboratory Press (1988).

Product List

Current prices may be obtained from our Web site or from our Customer Service Department.

Cat #	Product Name	Unit Size
A-6430	anti-dinitrophenyl-KLH, rabbit IgG fraction *2 mg/mL*	0.5 mL
A-11097	anti-dinitrophenyl-KLH, rabbit IgG fraction, Alexa Fluor® 488 conjugate *2 mg/mL*	0.5 mL
A-6435	anti-dinitrophenyl-KLH, rabbit IgG fraction, biotin-XX conjugate *2 mg/mL*	0.5 mL
A-6423	anti-dinitrophenyl-KLH, rabbit IgG fraction, fluorescein conjugate *2 mg/mL*	0.5 mL

Contact Information

Further information on Molecular Probes' products, including product bibliographies, is available from your local distributor or directly from Molecular Probes. Customers in Europe, Africa and the Middle East should contact our office in Leiden, the Netherlands. All others should contact our Technical Assistance Department in Eugene, Oregon.

Please visit our Web site — www.probes.com — for the most up-to-date information

Molecular Probes, Inc.

PO Box 22010, Eugene, OR 97402-0469
Phone: (541) 465-8300 • Fax: (541) 344-6504

Customer Service: 7:00 am to 5:00 pm (Pacific Time)

Phone: (541) 465-8338 • Fax: (541) 344-6504 • order@probes.com

Toll-Free Ordering for USA and Canada:

Order Phone: (800) 438-2209 • Order Fax: (800) 438-0228

Technical Assistance: 8:00 am to 4:00 pm (Pacific Time)

Phone: (541) 465-8353 • Fax: (541) 465-4593 • tech@probes.com

Molecular Probes Europe BV

PoortGebouw, Rijnsburgerweg 10
2333 AA Leiden, The Netherlands
Phone: +31-71-5233378 • Fax: +31-71-5233419

Customer Service: 9:00 to 16:30 (Central European Time)

Phone: +31-71-5236850 • Fax: +31-71-5233419

eurorder@probes.nl

Technical Assistance: 9:00 to 16:30 (Central European Time)

Phone: +31-71-5233431 • Fax: +31-71-5241883

eurotech@probes.nl

Molecular Probes' products are high-quality reagents and materials intended for research purposes only. These products must be used by, or directly under the supervision of, a technically qualified individual experienced in handling potentially hazardous chemicals. Please read the Material Safety Data Sheet provided for each product; other regulatory considerations may apply.

Several of Molecular Probes' products and product applications are covered by U.S. and foreign patents and patents pending. Our products are not available for resale or other commercial uses without a specific agreement from Molecular Probes, Inc. We welcome inquiries about licensing the use of our dyes, trademarks or technologies. Please submit inquiries by e-mail to busdev@probes.com. All names containing the designation ® are registered with the U.S. Patent and Trademark Office.

Copyright 2001, Molecular Probes, Inc. All rights reserved. This information is subject to change without notice.