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Product Specification Sheet

Product: IRDye™800 Conjugated Affinity Purified anti-Hemagglutinin (HA) EPITOPE TAG (Rabbit)

Code: 600-432-384

Lot # 12874

Size: 0.1 mg

Physical State: Lyophilized

Antibody Concentration: 1.0 mg/ml (by UV absorbance at 280 nm)

Label: IRDye™800 (MW 1166.2)

Fluorochrome/Protein Ratio: 3.0 moles IRDye™800/mole of Rabbit IgG

Absorption Wavelength: 774 nm (in PBS)

Emission Wavelength: 800 nm

Buffer: 0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2

Stabilizer: 10 mg/ml BSA IgG and Protease free

Preservative: 0.01% (w/v) Sodium Azide

Background Information: Epitope tags are short peptide sequences that are easily recognized by tag-specific antibodies. Due to their small size, epitope tags do not affect the tagged protein's biochemical properties. Most often sequences encoding the epitope tag are included with target DNA at the time of cloning to produce fusion proteins containing the epitope tag sequence. This allows anti-epitope tag antibodies to serve as universal detection reagents for any tag containing protein produced by recombinant means. This means that anti-epitope tag antibodies are a useful alternative to generating specific antibodies to identify, immunoprecipitate or immunoaffinity purify a recombinant protein. The anti-epitope tag antibody is usually functional in a variety of antibody-dependent experimental procedures. Expression vectors producing epitope tag fusion proteins are available for a variety of host expression systems including bacteria, yeast, insect and mammalian cells. Rockland Immunochemicals produces anti-epitope tag antibodies against many common epitope tags including Myc, GST, GFP, 6X His, MBP, FLAG and HA. Rockland Immunochemicals also produces antibodies to other tags including FITC, Rhodamine (TRITC), DNP and biotin.

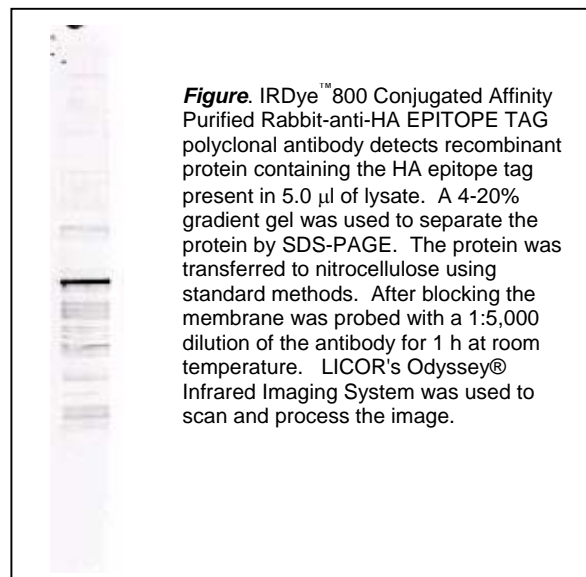


Figure. IRDye™800 Conjugated Affinity Purified Rabbit-anti-HA EPITOPE TAG polyclonal antibody detects recombinant protein containing the HA epitope tag present in 5.0 μ l of lysate. A 4-20% gradient gel was used to separate the protein by SDS-PAGE. The protein was transferred to nitrocellulose using standard methods. After blocking the membrane was probed with a 1:5,000 dilution of the antibody for 1 h at room temperature. LICOR's Odyssey® Infrared Imaging System was used to scan and process the image.

Application(s): Fluorescence technology is widely used to detect proteins. However, many common visible fluorophores often result in considerable background fluorescence in the visible range. Visible fluorophores are rarely used for membrane-based protein detection because of this high background. IRDye™ 800 antibody and reagent conjugates are specifically designed for protein detection methods that use longer-wavelength, near-infrared (IR) fluorophores to visualize proteins in western blotting and other applications. Very low background fluorescence in the IR range provides for a much higher signal-to-noise ratio than visible fluorophores. Detection levels in the picogram range on Western blots rival the sensitivity of chemiluminescence on film. IRDye™ 800 conjugates are optimized for the Odyssey® Infrared Imaging System developed by LI-COR. IRDye™ 800 conjugates are also suitable for immunofluorescence microscopy using commercially available excitation/emission filters in the 780nm/820nm range. Dual simultaneous labeling in western blots or microscopy is achieved when IRDye™ 800 conjugates are used in conjunction with Cy5.5™ conjugates. IRDye™ 800 conjugates provide an ultra-sensitive and convenient alternative to standard chemiluminescent protein detection methods, as well as a valuable tool for multicolor imaging. Anti-HA is optimally suited for monitoring the expression of HA tagged fusion proteins. As such, anti-HA/HA can be used to identify fusion proteins containing the HA epitope. The antibody recognizes the HA epitope tag fused to the amino- terminus of targeted proteins as is expressed in many commonly used expression vectors.

Recommended Dilution(s): This product was tested by immunoblot using antigen spotted to nitrocellulose membrane. A dilution range of 1:10,000 to 1:25,000 is suggested for this product to detect 12-25 pg of immobilized protein. Researchers should determine optimal titers for other applications.

Storage Conditions: Store vial at 4° C prior to restoration. Restore with 0.1 ml of deionized water (or equivalent). Centrifuge product if not completely clear after standing at room temperature. This product is stable at 4° C as an undiluted liquid. Dilute only prior to immediate use. Expiration date is one (1) year from date of restoration.

Purity and Specificity: This affinity purified antibody is directed against HA epitope tag and is useful in determining its presence in various assays. This polyclonal anti-HA tag antibody detects over-expressed proteins containing the HA epitope tag. To date this antibody has reacted with all HA tagged proteins so far tested. The antibody recognizes the HA-tag (Tyr-Pro-Tyr- Asp-Val-Pro-Asp-Tyr-Ala) fused to either the amino- or carboxy- termini of targeted proteins in transfected or transformed cells.

Immunogen: This antibody was purified from whole rabbit serum prepared by repeated immunizations with the 9-aa epitope tag peptide YPYDVPDYA (114-122) from hemagglutinin influenza conjugated to KLH using maleimide. A residue of cysteine was added to the carboxy terminal end to facilitate coupling.

Conjugation Reference: LI-COR Biosciences, Lincoln, NE.

Related Product(s):

#600-101-098	Affinity Purified Anti-BIOTIN (GOAT)
#600-401-382	Affinity Purified Anti-6X HIS TAG (Rabbit)
#600-101-200	Affinity Purified Anti-GST (GOAT)
#600-101-215	Affinity Purified Anti-GFP (GOAT)
#600-101-096	Affinity Purified Anti-FITC (GOAT)
#200-301-246	Protein A Purified Mouse Mab Anti-TRITC
#200-B01-380	Protein A Purified Hamster Mab Anti-DNP
#611-703-127	HRP Anti-Rabbit IgG [H&L] MX10 (DONKEY)
#611-132-122	IRDye800 Anti-Rabbit IgG [H&L] MX10 (GOAT)

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