

Certificate of Analysis

Product: IRDye™ 800CW Conjugated Affinity Purified anti-Goat IgG [H&L] [Donkey] Minimum Cross Reactivity to Chicken, Guinea Pig, Hamster, Horse, Mouse, Rabbit and Rat Serum Proteins

Code: 605-731-125

Lot # 20197

Size: 500 µg

Physical State: Lyophilized

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

Label: IRDye™ 800CW (MW 1166.2)

Fluorochrome/Protein Ratio: 1.5 moles IRDye™ 800CW/mole of Donkey 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

Application(s): IRDye™ 800CW conjugates are specifically designed for LI-COR's In-Cell Western assay for the Odyssey® Infrared Imaging System. IRDye™ 800CW conjugates can be used for western blotting applications, but the fluorochrome/protein ratio is not optimal for this application so detection sensitivity may be reduced. The In-Cell Western assay is a faster method for analyzing signal transduction pathways. In less time than a gel-based Western, you can quantify proteins in a 96- or 384-well microplate. In-Cell Westerns use infrared dye-labeled secondary antibodies to directly label proteins in fixed cultured cells, and quantify total fluorescence from each well. Time consuming and error-prone steps such as lysate preparation, gel loading and electrophoresis, and membrane transfer are eliminated with In-Cell Westerns. Simultaneous, two-target detection enables quantitative and accurate measurement of abundance or phosphorylation of one target by normalization against another target. Near-infrared probes yield high sensitivity for measuring small changes in protein amount or modification. Direct detection of proteins in their cellular context eliminates artifacts caused by cell lysis. The fast, microplate-based assay eliminates lysates, gels, and membranes required for conventional Western blots. The In-Cell Western assay is a moderate throughput method, ideal for screening cell treatments or drug candidates for their effects on target proteins. Recommended plates for In-Cell Westerns are Nunc-Nalgene (p/n 167008) for 96-well format and Falcon (BD Biosciences p/n 353961) for 384-well format. Confirm specificity and performance in the infrared system of primary and secondary antibodies by Odyssey western blotting *before* performing the In-Cell Western assay.

Recommended Dilution(s):

| | |
|--------------------------------|-----------------|
| LI-COR Odyssey® BLOT | Not Recommended |
| LI-COR In-Cell Western® | 1:800 - 1:1,200 |
| OTHER APPLICATIONS | User Optimized |

Storage Conditions: Store vial at 4° C prior to restoration. Restore with 0.5 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: This product was prepared from monospecific antiserum by immunoaffinity chromatography using Rat IgG coupled to agarose beads followed by solid phase adsorption(s) to remove any unwanted reactivities. Assay by immunoelectrophoresis resulted in a single precipitin arc against anti-Donkey Serum, Goat IgG and Goat Serum. No reaction was observed against Chicken, Guinea Pig, Hamster, Horse, Mouse, Rabbit and Rat Serum Proteins.

Immunogen: Goat IgG, whole molecule

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

Note: This material is subject to proprietary rights and is sold under license from LI-COR, Inc. This product is licensed for sale only for 'research-use' only. There is no implied license hereunder for any commercial use. IRDye is a trademark of LI-COR, Inc. COMMERCIAL USE shall include:

1. Resale, lease, license or other transfer of the material or any material derived or produced from it.
2. Resale, lease, license or other grant of rights to use this material or any material derived or produced from it.
3. Use of this material to perform services for a fee for third parties.

If you require a commercial license to use this material and do not have one, return this material, unopened to Rockland Inc. PO BOX 326, Gilbertsville, PA and money paid for the material will be refunded.