

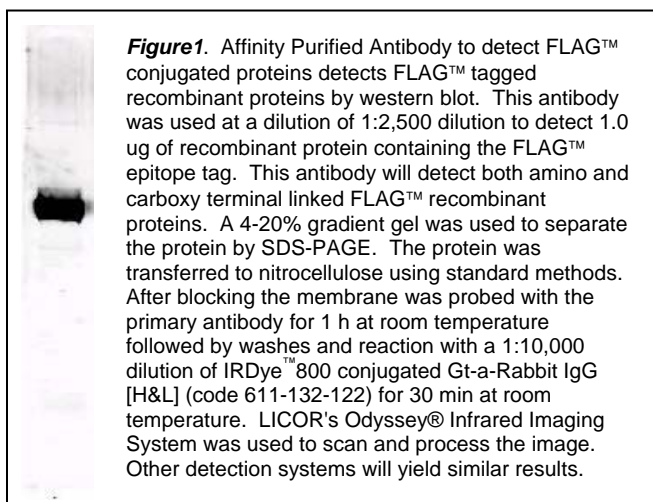


800-656-7625

fax 610-367-7825

**Certificate of Analysis****Product:** Affinity Purified Antibody for the detection of FLAG™™ conjugated proteins (Rabbit)**Code:** 600-401-383**Lot #:** 17976**Size:** 250 µg**Physical State:** Liquid (sterile filtered)**Antibody Concentration:** 0.71 mg/ml (by UV absorbance at 280 nm)**Buffer:** 0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2**Stabilizer:** None**Preservative:** 0.01% (w/v) Sodium Azide**Storage Conditions:** Store vial at -20° C. For extended storage aliquot contents and freeze at -20° C or below. Avoid cycles of freezing and thawing. Dilute only prior to immediate use. Expiration date is one (1) year from date of opening.

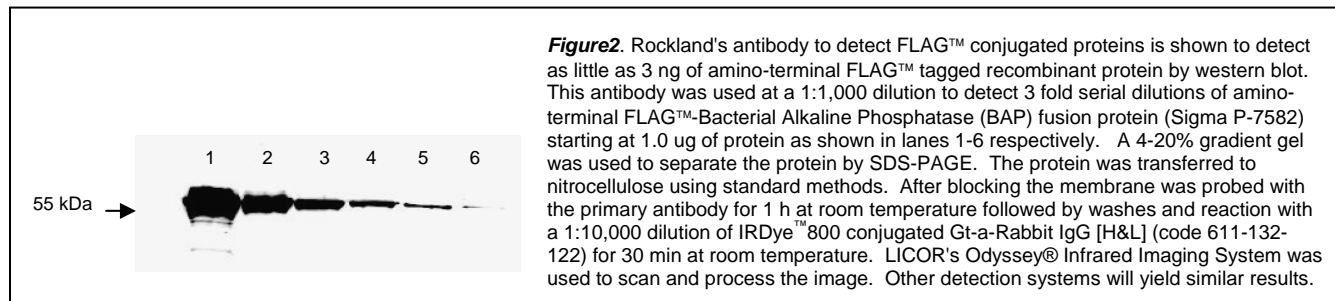
**Background Information:** Epitope tags are short peptide sequences that are easily recognized by tag-specific antibodies. Due to their small size, epitope tags do not affect the tagged protein's biochemical properties. Most often sequences encoding the epitope tag are included with target DNA at the 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.



<b>Recommended Dilutions:</b>	<b>ELISA</b>	1:20,000 - 1:100,000
	<b>WESTERN BLOT</b>	1:2,000 - 1:10,000
	<b>IMMUNOHISTOCHEMISTRY</b>	User Optimized
	<b>OTHER APPLICATIONS</b>	User Optimized

**Application Note(s):** This antibody is optimally suited for monitoring the expression of FLAG™ tagged fusion proteins. As such, this antibody can be used to identify fusion proteins containing the FLAG™ epitope. The antibody recognizes the epitope tag fused to either the amino- or carboxy termini of targeted proteins. This antibody has been tested by ELISA and western blotting against both the immunizing peptide and FLAG™ containing recombinant proteins. Although not tested, this antibody is likely functional for immunoprecipitation and immunocytochemistry, and other immunodetection techniques. The epitope tag peptide sequence was first derived from the 11-amino-acid leader peptide of the gene-10 product from bacteriophage T7. Now the most commonly used hydrophilic octapeptide is DYKDDDDK. Rockland Immunochemical's polyclonal antibody to detect FLAG™ conjugated proteins binds FLAG™ containing fusion proteins with greater affinity than the widely used monoclonal M1, M2 and M5 clones, and shows greater sensitivity in most assays. Affinity purification of the polyclonal antibody results in very low background levels in assays and low cross-reactivity with other cellular proteins.

**Purity and Specificity:** This affinity-purified antibody is directed against the FLAG™ motif and is useful in determining its presence in various assays. This polyclonal anti-FLAG™ tag antibody detects over-expressed proteins containing the FLAG™ epitope tag. In western blotting of bacterial extracts the antibody does not cross-react with endogenous proteins.



**Immunogen:** This antibody was purified from whole rabbit serum prepared by repeated immunizations with the Enterokinase Cleavage Site (ECS) peptide DYKDDDDK (Asp-Tyr-Lys-Asp-Asp-Asp-Lys) conjugated to KLH using maleimide. Residues of glycine and cysteine were added to the carboxy terminal end to facilitate coupling. This antibody reacts with FLAG™ conjugated proteins.

#### Related Products:

<a href="#">#600-401-381</a>	Affinity Purified Anti-Myc TAG (Rabbit)
<a href="#">#600-401-382</a>	Affinity Purified Anti-6X His TAG (Rabbit)
<a href="#">#600-401-383</a>	Affinity Purified Antibody for the detection of FLAG™ conjugated proteins (Rabbit)
<a href="#">#600-431-383</a>	IRDYE800CW™ Conjugated Affinity Purified Antibody for the detection of FLAG™ conjugated proteins (Rabbit)
<a href="#">#600-432-383</a>	IRDYE800™ Conjugated Affinity Purified Antibody for the detection of FLAG™ conjugated proteins (Rabbit)
<a href="#">#600-401-384</a>	Affinity Purified Anti-HEMAGGLUTININ (Rabbit)
<a href="#">#200-101-150</a>	IgG Fraction of Anti-LUCIFERASE (GOAT)
<a href="#">#600-101-200</a>	Affinity Purified Anti-GST (GOAT)
<a href="#">#611-703-127</a>	Peroxidase Conjugated Affinity Purified Anti-RABBIT IgG (H&L) (DONKEY) MX10
<a href="#">#611-132-122</a>	IRDye800 Conjugated Affinity Purified Anti-RABBIT IgG (H&L) (GOAT) MX10
<a href="#">#MB-070</a>	Blocking Buffer for Fluorescent Western Blotting
<a href="#">#KIA-003</a>	<b>MaxTag™</b> Anti-RABBIT IgG Kit for Immunoblotting

#### General References:

- Chubet, R.G. and Brizzard, B.L. (1996) Vectors for expression and secretion of FLAG epitope-tagged proteins in mammalian cells. *Biotechniques* **20**(1):136-141.
- Slootstra, J.W. et al., (1997) Identification of new tag sequences with differential and selective recognition properties for the anti-FLAG monoclonal antibodies M1, M2 and M5. *Mol Divers* **2**(3):156-164.
- Robeva, A.S. et al., (1996) Double tagging recombinant A1- and A2A-adenosine receptors with hexahistidine and the FLAG epitope. Development of an efficient generic protein purification procedure. *J Biochem Pharmacol* **51**:4, 545-55.
- Fulton, J.E. et al., (1995) Functional analysis of avian class I (BIV) glycoproteins by epitope tagging and mutagenesis in vitro. *Eur J Immunol* **25**:7, 2069-76.

**USDA Certification:** All products of animal origin manufactured by Rockland Immunochemicals are derived from starting materials of North American origin. Collection was performed in United States Department of Agriculture (USDA) inspected facilities and all materials have been inspected and certified to be free of disease and suitable for exportation.

**Note:** This product is for research use only and is not intended for therapeutic or diagnostic applications. Please contact a technical service representative for more information.