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R1064PS Polyclonal Antibody to Beta-galactosidase tag - Purified

Alternate names: Beta-Gal Fusion Protein, Beta-Gal tag, JW0335, Lactase, b0344, lacZ tag

Quantity: 0.1 mg

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

Background: Beta-galactosidase is coded by a gene (lac z) in the lac operon of Escherichia coli. It is a metalloenzyme that splits lactose into glucose and galactose. It hydrolyzes terminal, non-reducing beta-D-galactose residues in beta-D-galactosides. Activation by cations seems to be substrate dependent. K^+, Na^+, NH_4^+, Rb^+, Cs+ and Mn^{++} all activate enzyme activity based upon the substrate used.
Anti Beta-galactosidase antibody recognizes the enzyme beta-galactosidase, or ß-galactosidase, that is a component of assays used frequently in genetics, molecular biology (see X-gal) for a blue white screen, and other life sciences. IPTG induces production of ß-galactosidase by binding and inhibiting the lac repressor. Since it is highly expressed and accumulated in lysosomes in senescent cells, it is used as a senescence biomarker both in vivo and in vitro in qualitative and quantitative assays, despite its limitations. Anti Beta-galactosidase antibody is ideal for investigators involved in enzyme research.

Uniprot ID: P00722

NCBI: AP_000996.1

GeneID: 945006

Host: Rabbit

Immunogen: Full length native Beta-galactosidase isolated from *E.coli*

Format: **State:** Lyophilized purified IgG fraction
Purification: Multi-step process which includes delipidation, salt fractionation and ion exchange chromatography followed by extensive dialysis against the buffer
Buffer System: 0.02M Potassium phosphate, 0.15M Sodium chloride, pH 7.2
Preservatives: 0.01% (w/v) Sodium azide
Stabilizers: None
Reconstitution: Restore with 0.1 ml of deionized water (or equivalent).

Applications: Suitable for Immunoblotting (Western or dot blot), ELISA, Immunofluorescence microscopy, Immunoprecipitation, conjugation and most immunological methods requiring high titer and specificity.
Western blot: 1/5,000-1/10,000. A 1/5,000 dilution has been reported to be successful for staining by Immunoblot of Beta-galactosidase fusion proteins after transfer using a semi-dry transfer apparatus). **ELISA**: 1/10,000.
Immunohistochemistry: 1/1,500. The antibody recognizes both Frozen tissue sections, Paraffin embedded tissue and 4% paraformaldehyde fixed tissue for most immunohistochemical analysis:A 1/1,500 dilution has been reported to detect Beta-galactosidase in adult rat spinal cord tissue after infection with helper-dependent adenovirus expressing lacZ. In this particular experiment, tissue was perfused with 4% paraformaldehyde and cryostat-cut (35 µm) to produce free-floating sections. A

1/5,000 dilution has been reported for immunofluorescent staining of methanol fixed, devitellinized Drosophila embryos. Although a wide range of conditions was reported to be effective, a 1/10,000 dilution was noted to show no background and to be suitable for double labeling experiments). A 1/5,000 dilution has been reported to be successful for staining brain sections from transgenic mice expressing nuclear Beta-galactosidase when assayed by **Immunofluorescence microscopy**.
Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.

Specificity:

This antibody detects Beta-galactosidase [*E. coli*]. Cross reactivity against Beta-galactosidase from other tissues and species may occur but have not been specifically determined. Very low background staining has been reported in various assays.
Immunoelectrophoresis gives a single precipitin arc against anti-rabbit serum as well as purified and partially purified Beta-galactosidase *[E.coli]*.

Add. Information:

Conjugates available. Please ask for details.

Storage:

Store lyophilized at 2-8°C for 6 months or at -20°C long term.
After reconstitution store the antibody undiluted at 2-8°C for one month or (in aliquots) at -20°C long term. Dilute only prior to immediate use. Avoid repeated freezing and thawing.
Shelf life: one year from despatch.

Product Citations:

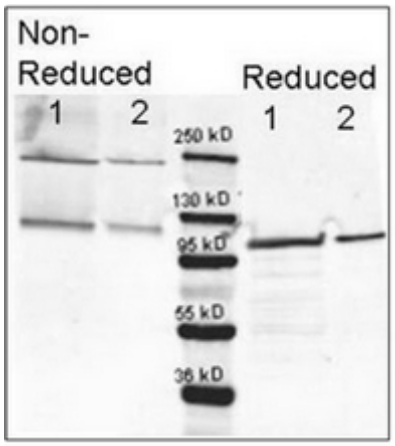
Originator or purchased from resellers:
1. Hayworth CR, Moody SE, Chodosh LA, Krieg P, Rimer M, Thompson WJ. Induction of neuregulin signaling in mouse schwann cells in vivo mimics responses to denervation. J Neurosci. 2006 Jun 21;26(25):6873-84. PubMed PMID: 16793894.

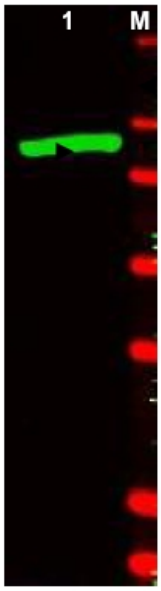
General Readings:

1. Huber RE, Hakda S, Cheng C, Cupples CG, Edwards RA. Trp-999 of beta-galactosidase (Escherichia coli) is a key residue for binding, catalysis, and synthesis of allolactose, the natural lac operon inducer. Biochemistry. 2003 Feb 18;42(6):1796-803. PubMed PMID: 12578395.
2. Matthews BW. The structure of E. coli beta-galactosidase. C R Biol. 2005 Jun;328(6):549-56. PubMed PMID: 15950161.

Pictures:

Western blotting using Beta-galactosidase antibody Cat.-No R1064PS. Lane 1 shows 80 ng and lane 2 shows 20 ng loaded onto gel. Results for Non-Reducing Conditions of SDS-PAGE prior to transfer to nitrocellulose are shown on the left side of the figure; results obtainined under Reducing Conditions are shown on the right. Blots were blocked overnight at 4°C with blocking buffer for fluorescent Western blotting (p/n MB-070). The membrane was probed with Beta-galactosidase antibody Cat.-No R1064P diluted to 1/10,000. Reaction occurred overnight at 4°C. Dylight649™ conjugated goat-anti-rabbit IgG was used for detection. Molecular weight estimation was made by comparison to a prestained MW marker (center). Fluorescence image was captured using the VersaDoc® imaging system developed by BIO-RAD. Other detection systems will yield similar results.





Western blot using Beta-galactosidase antibody Cat.-No. R1064PS shows detection of a band at ~117 kDa (lane 1) corresponding to the protein present in partially purified preparations. Approximately 50 ng of protein was separated on a 4-20% Tris-Glycine gel by SDSPAGE and transferred onto nitrocellulose. After blocking the membrane was probed with the primary antibody diluted to 1/1,000. Reaction occurred overnight at 4°C followed by washes and reaction with a 1/10,000 dilution of IRDye800(TM) conjugated goat-a-rabbit IgG [H&L] for 45 min at RT (800 nm channel, green). Molecular weight estimation was made by comparison to prestained MW markers in lane M (700 nm channel, red). RDye800(TM) fluorescence image was captured using the Odyssey(R) infrared imaging system developed by LI-COR. IRDye is a trademark of LI-COR, Inc. Other detection systems will yield similar results.