Rabbit anti-Human 3'Sulfo Lewis X Antibody

Product code: B4 Rb IgG -50

Clone name: E2-B4

Product type: Recombinant rabbit IgG monoclonal antibody

Lot number: T2234A23

Source: Cell culture expression in HEK293 cells

Purification: Purified by Protein A or Protein G affinity chromatography

Immunogen: 3'Sulfo-Lewis X

Buffer: Phosphate buffered saline pH7.5

Endotoxin: <0.050 EU/mg as determined by LAL chromogenic endotoxin assay

Concentration: 1mg/ml

Vial size: 0.05mg

Target information:

This antibody recognises the 3'sulfo-βGal linkage found on the tetrasaccharide carbohydrate Sialyl LewisX (sLeX), usually found attached to O-glycans on the surface of cells. 3'-Sulfo-Lewis X (3-SLeX) is a glycan epitope that has been reported to act as a ligand for the adhesion molecules E- and L-selectin. The synthesis of 3-SLeX is mediated by the enzyme galactose-3-O-sulfotransferase (GAL3ST2), which is expressed in cell lines with high metastatic potential.

Expression of 3-SLeX is therefore potentially relevant to the identification of tumours with poor prognosis and higher likelihood of progression and metastasis.

3-SLeX has been found to be expressed at high levels in the mucins from the lungs of Cystic Fibrosis patients. It has been shown that Pseudomonas Aeruginosa preferentially binds to this form of LeX, and that high levels of expression may be related to more serious infections and chronic colonisation of airway mucins in Cystic Fibrosis.

Studies of 3-SLeX expression and function have often been inhibited by the lack of any specific antibodies. Glykogen have overcome these reagent challenges by using their proprietary immunisation methods to develop Clone B4, a recombinant monoclonal rabbit IgG antibody. The availability of this antibody will facilitate studies into 3-SLeX expression and function, both in normal tissues and in cancer studies.

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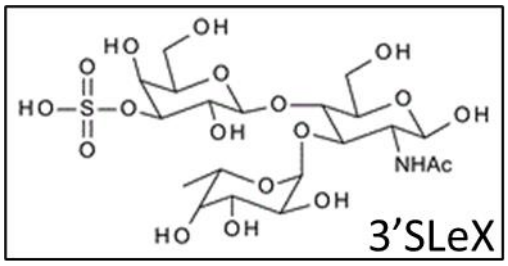
The chemical structure of the 3-SLeX antigen is shown below

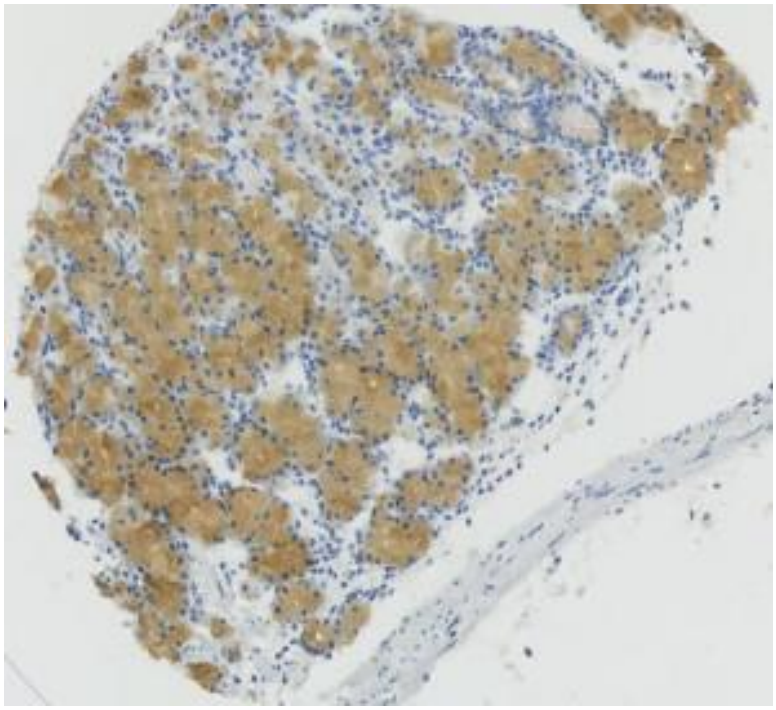


Applications:

Immunohistochemistry: This antibody may be used to stain both frozen and paraffin embedded tissue sections.

For staining of paraffin sections antigen retrieval using a TrisEDTA buffer, pH9, is recommended. A dilution range of 10-40 ug/ml is suggested, but users should titrate the antibody for best results within their own systems.

The image below shows staining FFPE oesophageal adenocarcinoma tissue.



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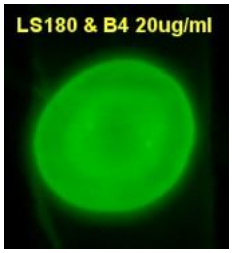
Immunoblotting: This antibody may be used for immunoblotting of lipid extracts of cells. A dilution range of 10-50 ug/ml is suggested, but users should titrate the antibody for best results within their own systems. NB: Due to the glycan nature of the epitope recognised by this antibody, traditional protein Western Blotting cannot be used.

The image below shows lipid immunoblotting of a total lipid extract of LS180 cells using the B4 antibody at 20ug/ml (with Sigma A0545 Goat anti-rabbit IgG peroxidase).



ELISA: This antibody may be used in ELISA applications. A dilution range of 20-50 ug/ml is suggested, but users should titrate the antibody for best results within their own systems

Recommended secondary antibody: A suitable anti-rabbit IgG reagent such as product A0545 (Goat anti-rabbit IgG:HRP) or SAB3700963 (Donkey anti-rabbit IgG (H+L) $F(ab')_2$:FITC from Sigma is recommended

Shipping: The product is shipped at ambient temperature. Upon receipt store immediately as recommended below

Storage: Use a manual defrost freezer and avoid repeated freeze/thaw cycles. Store frozen in aliquots at -20^0C for up to one year, or at 4^0C for up to one week.

References

1. Yuen CT, Bezouska K, O'Brien J, et al. Sulfated blood group Lewisa. A superior oligosaccharide ligand for human E-selectin. *Journal of Biological Chemistry*. 1994;269(3):1595-1598. doi:10.1016/s0021-9258(17)42065-5

2.Green PJ, Tamatani T, Watanabe T, et al. High affinity binding of the leucocyte adhesion molecule L-selectin to 3'-sulphated-Lea and -Lex oligosaccharides and the predominance of sulphate in this interaction demonstrated by binding studies with a series of lipid-linked oligosaccharides. *Biochem Biophys Res Commun*. 1992;188(1). doi:10.1016/0006-291X(92)92376-9

3. Yuen CT, Stoll MS, Feizi T, et al. Novel Sulfated Ligands for the Cell Adhesion Molecule E-Selectin Revealed by the Neoglycolipid Technology among O-Linked Oligosaccharides on an Ovarian Cystadenoma Glycoprotein. *Biochemistry*. 1992;31(38):9126-9131. doi:10.1021/bi00153a003

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