

Recombinant Human Glypican 3 His-tag Alexa Fluor® 647

Catalog Number: AFR2119

DESCRIPTION	
Source	Mouse myeloma cell line, NS0-derived human Glypican 3 protein Gln25-His559 with a C-terminal 6-His tag Accession # P51654.1
N-terminal Sequence Analysis	No results obtained: Gln25 predicted, Ser359 & Val483
Structure / Form	Glypican 3 is subject to endoproteolytic processing by proprotein convertases (PC). By amino acid sequencing, three peptides (the first with a blocked N-terminus most likely starts with Gln25, the second peptide starts with Ser359 after a furin cleavage site, and the third peptide starts with Val483) are present in the recombinant GPC3 preparation. Peptides 2 and 3 are detected at a 1:1 ratio. All three peptides remained associated via disulfide bonds. Labeled with Alexa Fluor® 647 Excitation Wavelength: 650 nm Emission Wavelength: 668 nm
Predicted Molecular Mass	62 kDa

SPECIFICATIONS	
SDS-PAGE	60-100 kDa, under non-reducing conditions.
Activity	Measured by flow cytometry for its ability to bind anti-human Glypican 3 Antibody conjugated beads. The concentration of Recombinant Human Glypican 3 His-tag Alexa Fluor® 647 (Catalog # AFR2119) that produces 50% of the binding response is 0.500-10.0 ng/mL.
Endotoxin Level	<1.0 EU per 1 μ g of the protein by the LAL method.
Purity	>95%, by SDS-PAGE visualized with Silver Staining and quantitative densitometry by Coomassie® Blue Staining.
Formulation	Supplied as a 0.2 µm filtered solution in PBS with BSA as a carrier protein. See Certificate of Analysis for details.

PREPARATION AND STORAGE	
Shipping	The product is shipped with dry ice or equivalent. Upon receipt, store it immediately at the temperature recommended below.
Stability & Storage	Protect from light. Use a manual defrost freezer and avoid repeated freeze-thaw cycles.
	 6 months from date of receipt, -20 to -70 °C as supplied.
	 1 month, 2 to 8 °C under sterile conditions after opening.

- 3 months, -20 to -70 °C under sterile conditions after opening.



Flow cytometry analysis for Recombinant Human Glypican 3 His-tag Alexa Fluor® 647 staining on Anti-Human Glypican 3 Antibody conjugated beads. Streptavidin coated beads conjugated to biotinylated Anti-Human Glypican 3 (Catalog # BAF2119) were stained with the indicated concentrations of Recombinant Human Glypican 3 His-tag Alexa Fluor® 647 (Catalog # AFR2119).

Rev. 6/22/2022 Page 1 of 2



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BACKGROUND

Glypicans (GPC) are a family of heparan sulfate proteoglycans that are attached to the cell surface by a glycosylphosphatidylinositol (GPI) anchor. Six members of this family have been identified in mammals (GPC1-GPC6). All glypican core proteins contain an N-terminal signal peptide, a large globular cysteine-rich domain (CRD) with 14 invariant cysteine residues, a stalk-like region containing the heparan sulfate attachment sites, and a C-terminal GPI attachment site. While glypican proteins do not share strong amino acid sequence identity (they range from 17-63%), the conserved cysteine residues in their CRDs suggests similarity in their three-dimensional structure (1, 2). Mutations in GPC3 cause a rare disorder in humans, Simpson-Golabi-Behmel Syndrome, which is characterized by pre and postnatal overgrowth of multiple tissues and organs and an increased risk for developing embryonic tumors (3). These features are also present in the mouse knock-out of GPC3 indicating that GPC3 regulates cell survival and inhibits cell proliferation during development (4). Glypican 3 has been implicated in regulating many different signaling pathways including: IGF, FGF, BMP and Wnt. An endoproteolytic processing of GPC3 by proprotein convertases is required for the modulation of Wnt signaling (5). Direct interaction with FGF-basic has been observed and is mediated by the heparan sulfate chains (6).

References:

- 1. Filmus, J. and S.B. Selleck (2001) J. Clinical Invest. 108:497.
- 2. De Cat, B and G. David (2001) Seminars in Cell & Dev. Biol. 12:117.
- 3. Pilia, G. et al. (1996) Nat. Genet. 12: 241.
- 4. Cano-Gauci, D.F. et al. (1999) J. Cell Biol. 146: 255.
- 5. De Cat, B. *et al.* (2003) J. Cell Biol. **163**:625.
- 6. Song, H.H. et al. (1997) J. Biol. Chem. 272:7574

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Rev. 6/22/2022 Page 2 of 2



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