

EPHB1

aa565-984

Product No.: 0317-0000-1

Lot: 001

Description: Human EPHB1

Amino acids R₅₆₅-A₉₈₄ (as in GenBank entry NM_004441.2)*, N-terminally fused to GST-HIS₆-Thrombin cleavage site

*Sequence may contain documented polymorphisms
Detailed sequence on request

Product identity: EPHB1, Lot 001, was confirmed as human EPHB1 by mass spectroscopy LC-ESI-MS/MS (Protagen AG, Germany)

Theoretical MW_{Fusion Protein}: 76,906 Da

Expression: Baculovirus infected Sf9 cells

Purification: One-step affinity purification using GSH-agarose

Storage buffer: 50 mM Tris-HCl, pH 8.0; 100 mM NaCl, 5 mM DTT, 15 mM reduced glutathione, 20% glycerol

Storage temperature: -80°C
Avoid repeated freeze-thaw cycles!

Protein concentration: 0.500 µg/µl
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

Method for determination of K_m value & Specific activity:

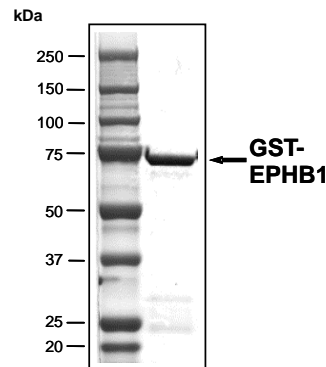
• Assay conditions:

- 60 mM HEPES-NaOH, pH 7.5
- 3 mM MgCl₂
- 3 mM MnCl₂
- 3 µM Na-orthovanadate
- 1.2 mM DTT
- 2.5 µg / 50 µl PEG_{20,000}
- ATP (variable)
- Substrate: Poly(Glu,Tyr)_{4:1}
(Sigma P-0275), 1 µg / 50 µl
- Recombinant EPHB1: 50 ng / 50 µl

- Filter binding assay
MAFC membrane (Millipore)

Specific activity: 72 pmol/µg×min

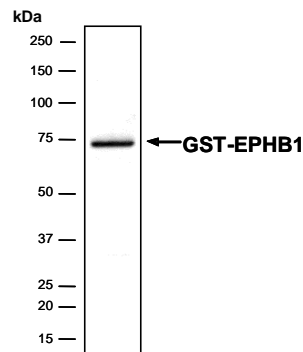
Coomassie stain:



2.0 µg GST-EPHB1

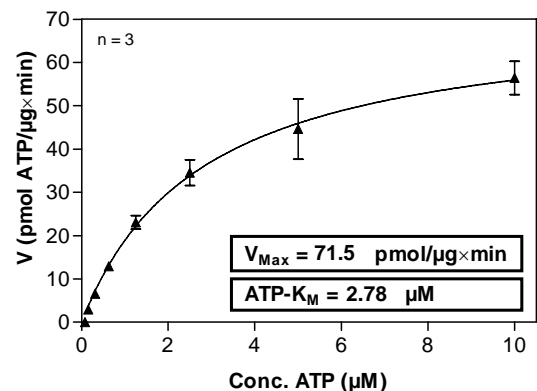
Western blot analysis:

anti-GST



0.5 µg GST-EPHB1

Determination of K_m value for ATP:



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