



#7486

EPHA3

aa569-983

Product No.: 0351-0000-1

Description: Human EPHA3

Amino acids G₅₆₉-V₉₈₃ (as in GenBank entry NM_005233)*, N-terminally fused to GST-HIS₆-Thrombin cleavage site

*Sequence may contain documented polymorphisms
Detailed sequence on request

Product identity: EPHA3, Lot 001, was confirmed as human EPHA3 by mass spectroscopy LC-ESI-MS/MS (Protagen AG, Germany)

Theoretical MW_{Fusion Protein}: 75,680 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, 4 mM reduced glutathione, 20% glycerol

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

Protein concentration: 0.140 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 EPHA3: 200 ng / 50 µl

• Filter binding assay MSFC membrane (Millipore)

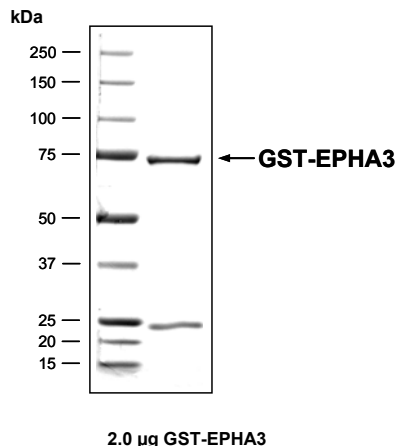
Specific activity: 184 pmol/µg×min

This enzyme was purified and extensively validated in a radioactive filter binding assay by a collaborator of Cell Signaling Technology. Cell Signaling Technology is in the process of revalidating this reagent in house using several technologies relevant to the screening community including antibody based detection methods.

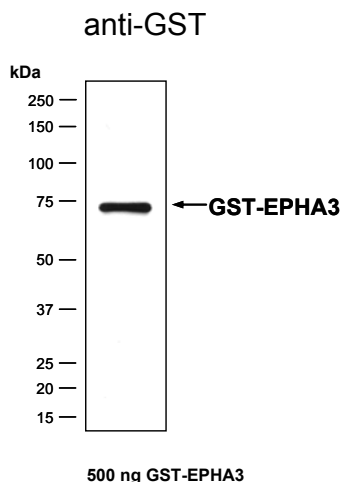
Please contact us at drugdiscovery@cellsignal.com for details.

This product is for *in vitro* research use only and is not intended for use in humans or animals.

Coomassie stain:



Western blot analysis:



Determination of K_m value for ATP:

