Anti-PDGFR alpha (phospho Y754) antibody ab5460

**Overview**

**Product name**
Anti-PDGFR alpha (phospho Y754) antibody

**Description**
Rabbit polyclonal to PDGFR alpha (phospho Y754)

**Host species**
Rabbit

**Tested applications**
Suitable for: ICC/IF, IHC-P, WB

**Species reactivity**
Reacts with: Mouse, Human

**Predicted to work with:** Rat, Xenopus laevis

**Immunogen**
Synthetic peptide corresponding to Human PDGFR alpha.
(Peptide available as ab5695)

**Positive control**
NIH3T3 cells +/- PDGF.

**Properties**

**Form**
Liquid

**Storage instructions**
Shipped at 4°C. Upon delivery aliquot and store at -20°C or -80°C. Avoid repeated freeze / thaw cycles.

**Storage buffer**
Preservative: 0.05% Sodium azide
Constituents: PBS, 0.1% BSA

BSA is IgG and protease free

**Purity**
Immunogen affinity purified

**Purification notes**
The antibody has been negatively preadsorbed using a non-phosphopeptide corresponding to the site of phosphorylation to remove antibody that is reactive with non-phosphorylated PDGFR alpha. The final product is generated by affinity chromatography using a PDGFR alpha derived peptide phosphorylated at tyrosine 754.

**Clonality**
Polyclonal

**Isotype**
IgG

**Applications**

Our Abpromise guarantee covers the use of ab5460 in the following tested applications.
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

<table>
<thead>
<tr>
<th>Application</th>
<th>Abreviews</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>ICC/IF</td>
<td></td>
<td>Use at an assay dependent concentration. 1/50 (see Abreview)</td>
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<tr>
<td>IHC-P</td>
<td></td>
<td>1/20.</td>
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<tr>
<td>WB</td>
<td></td>
<td>Use a concentration of 0.35 - 1 µg/ml. Detects a band of approximately 170-185 kDa.</td>
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**Target**

**Function**
Receptor that binds both PDGFA and PDGFB and has a tyrosine-protein kinase activity.

**Tissue specificity**
Expressed in primary and metastatic colon tumors and in normal colon tissue. Tumors may express a different isoform to that found in normal tissue.

**Involvement in disease**
Note=A chromosomal aberration involving PDGFRA is found in some cases of hypereosinophilic syndrome. Interstitial chromosomal deletion del(4)(q12q12) causes the fusion of FIP1L1 and PDGFRA (FIP1L1-PDGFRA).

**Sequence similarities**
Belongs to the protein kinase superfamily. Tyr protein kinase family. CSF-1/PDGF receptor subfamily. Contains 5 Ig-like C2-type (immunoglobulin-like) domains. Contains 1 protein kinase domain.

**Cellular localization**
Membrane.

**Images**

Immunohistochemical analysis of Paraffin embedded mouse colon tissue labeling PDGFR alpha (phospho Y754) with ab5460 (right) compared to a negative control section without primary antibody (left).

Heat mediated antigen retrieval was performed using 10mM sodium citrate (pH 6.0). The secondary was a HRP-conjugated secondary antibody.
Peptide Competition: Extracts prepared from NIH3T3 cells left unstimulated (1) and stimulated with PDGF (2-5) were resolved by SDS-PAGE on a 10% polyacrylamide gel and transferred to PVDF. Membranes were blocked with a 5% BSA-TBST buffer overnight at 4°C, then were incubated with 0.50 µg/mL ab5460 antibody for two hours at room temperature in a 1% BSA-TBST buffer, following prior incubation with: no peptide (1, 2), the non-phosphopeptide corresponding to the immunogen (3), a generic phosphotyrosine containing peptide (4), or, the phosphopeptide immunogen (5). After washing, membranes were incubated with goat F(ab')2 anti-rabbit IgG alkaline phosphatase and bands were detected using the Tropix WesternStar method. The data show that only the peptide corresponding to ab5460 blocks the antibody signal, thereby demonstrating the specificity of the antibody.

Immunocytochemistry/Immunofluorescence analysis of human glioblastoma cells labeling PDGFR alpha (phospho Y754) with ab5460 at 1/50 dilution. A goat anti-rabbit FITC conjugated antibody was used as the secondary antibody (1/400). Cells were fixed in paraformaldehyde and permeabilized with 0.1% Triton X-100.
Immunohistochemical analysis of Paraffin embedded human brain tissue labeling PDGFR alpha (phospho Y754) with ab5460 (right) compared to a negative control section without primary antibody (left).

Heat mediated antigen retrieval was performed using 10mM sodium citrate (pH 6.0). The secondary was a HRP-conjugated secondary antibody.

ab5460 staining PDGFR alpha (phospho Y754) in murine brain tissue by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections). Tissue was fixed with paraformaldehyde and a heat mediated antigen retrieval step was performed using citric acid pH 6.1. Samples were then permeabilized using 0.3% H$_2$O$_2$, then blocked with 0.5% BSA for 20 minutes at room temperature, followed by incubation with the primary antibody at a 1/50 dilution for 1 hour. An undiluted HRP-conjugated Goat anti-rabbit/mouse polyclonal was used as secondary antibody. Nuclei staining with Haematoxilin (blue) and PDGFR Receptor alpha staining with DAB (brown). PDGFR alpha staining in the membrane of astrocytes.

ab5460 staining PDGFR alpha (phospho Y754) in human glioblastoma multiforme brain tissue by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections). Tissue was fixed with paraformaldehyde and a heat mediated antigen retrieval step was performed using citric acid pH 6.1. Samples were then permeabilized using 0.3% H$_2$O$_2$, then blocked with 0.5% BSA for 20 minutes at room temperature, followed by incubation with the primary antibody at a 1/50 dilution for 1 hour. An undiluted HRP-conjugated Goat anti-rabbit/mouse polyclonal was used as secondary antibody. Nuclei staining with Haematoxilin (blue) and PDGFR alpha staining with DAB (brown). PDGFR alpha staining in the membrane of astrocytes.

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