abcam

Product datasheet

Anti-Alkaline Phosphatase, Tissue Non-Specific antibody [2F4] ab126820

KO VALIDATED

7 References 7 Images

Overview

Product name Anti-Alkaline Phosphatase, Tissue Non-Specific antibody [2F4]

Description Mouse monoclonal [2F4] to Alkaline Phosphatase, Tissue Non-Specific

Host species Mouse

Tested applications Suitable for: WB, ELISA, IHC-P, Flow Cyt, ICC/IF

Species reactivity Reacts with: Human

Immunogen Recombinant fragment corresponding to Human Alkaline Phosphatase, Tissue Non-Specific.

expressed in E.coli
Database link: **P05186**

Positive control Recombinant Human ALPL protein (AA: 18-502), HeLa and NTERA-2 cell lysates. This antibody

gave a positive result when used in the following formaldehyde/methanol fixed cell lines: HepG2

General notesThis product was changed from ascites to supernatant. Lot no's high than GR307490-5 are from

Tissue Culture Supernatant

The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at -20°C.

Storage buffer Preservative: 0.05% Sodium azide

Constituent: 99% PBS

Purity Protein G purified

Purification notes Purified from tissue culture supernatant.

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Clonality Monoclonal

Clone number 2F4
Isotype IgG1

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab126820 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		1/500 - 1/2000. Detects a band of approximately 57.3, 78.9 kDa (predicted molecular weight: 57, 78.9 kDa).
ELISA		1/10000.
IHC-P		1/200 - 1/1000.
Flow Cyt		1/200 - 1/400. ab170190 - Mouse monoclonal lgG1, is suitable for use as an isotype control with this antibody.
ICC/IF		1/100.

Target

Function This isozyme may play a role in skeletal mineralization.

Involvement in disease Hypophosphatasia

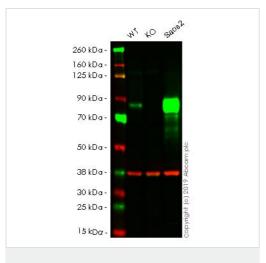
Hypophosphatasia childhood type Hypophosphatasia infantile type

Sequence similarities Belongs to the alkaline phosphatase family.

Post-translational N-glycosylated. modifications

Cellular localization Cell membrane.

Images



Western blot - Anti-Alkaline Phosphatase, Tissue Non-Specific antibody [2F4] (ab126820)

All lanes : Anti-Alkaline Phosphatase, Tissue Non-Specific antibody [2F4] (ab126820) at 1/500 dilution

Lane 1 : Wild-type HAP1 whole cell lysate

Lane 2: ALPL knockout HAP1 whole cell lysate

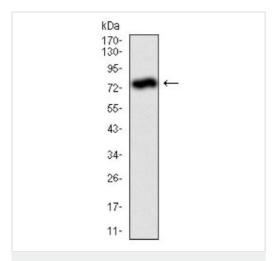
Lane 3: Saos2 whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 57, 78.9 kDa

Lanes 1 - 3: Merged signal (red and green). Green - ab126820 observed at 80 kDa. Red - loading control, <u>ab181602</u>, observed at 37 kDa.

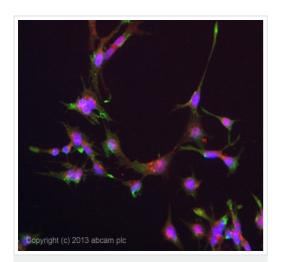
ab126820 was shown to specifically react with ALPL in wild-type HAP1 cells as signal was lost in ALPL knockout cells. Wild-type and ALPL knockout samples were subjected to SDS-PAGE. The membrane was blocked with 3% Milk. Ab126820 and ab181602 (Rabbit anti-GAPDH loading control) were incubated overnight at 4°C at 1/500 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed ab216772 and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preabsorbed ab216777 secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-Alkaline Phosphatase, Tissue Non-Specific antibody [2F4] (ab126820)

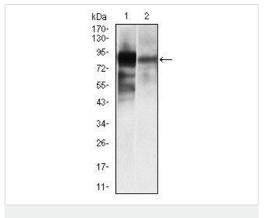
Anti-Alkaline Phosphatase, Tissue Non-Specific antibody [2F4] (ab126820) at 1/500 dilution + Recombinant Human protein ALPL (AA: 18-502)

Predicted band size: 57, 78.9 kDa

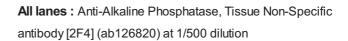


Immunocytochemistry/ Immunofluorescence - Anti-Alkaline Phosphatase, Tissue Non-Specific antibody [2F4] (ab126820)

ICC/IF image of ab126820 stained HepG2 cells. The cells were 4% formaldehyde fixed (10 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody ab126820 at 1/100 dilution overnight at +4°C. The secondary antibody (green) was DyLight® 488 goat anti- mouse (ab96879) lgG (H+L) used at a 1/250 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43 μ M. This antibody also gave a positive result in methanol fixed (100%, 5min) hepG2 cells used at 1/100 dilution.

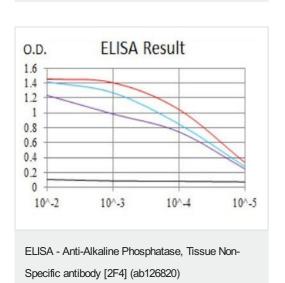


Western blot - Anti-Alkaline Phosphatase, Tissue Non-Specific antibody [2F4] (ab126820)

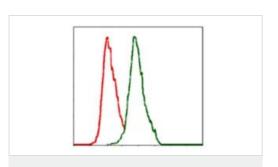


Lane 1 : HeLa cell lysate
Lane 2 : NTERA-2 cell lysate

Predicted band size: 57, 78.9 kDa

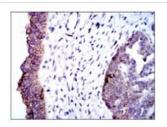


Example of standard curves obtained using ab126820. Control antigen 100ng (black), antigen at 10ng (purple), antigen at 50ng (blue), antigen at 100ng (red)



Flow Cytometry - Anti-Alkaline Phosphatase, Tissue Non-Specific antibody [2F4] (ab126820)

Flow cytometric analysis of MCF-7 cells using ab126820 at 1/200 (green) and negative control (red).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Alkaline Phosphatase,
Tissue Non-Specific antibody [2F4] (ab126820)

ab126820, at 1/200, staining Alkaline Phosphatase, Tissue Non-Specific in paraffin embedded Human ovarian cancer tissues by Immunohistochemistry with DAB staining.

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