Overview

Product name: Anti-Osteoprotegerin antibody
Description: Rabbit polyclonal to Osteoprotegerin
Host species: Rabbit
Tested applications: Suitable for: ICC/IF, IHC-P, WB
Species reactivity: Reacts with: Rat, Human
Predicted to work with: Mouse, Sheep, Cow, Pig

Immunogen: Synthetic peptide conjugated to KLH derived from within residues 200 - 300 Osteoprotegerin. Read Abcam's proprietary immunogen policy (Peptide available as ab73399.)


Properties

Form: Liquid
Storage instructions: Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -80°C. Avoid freeze / thaw cycle.
Storage buffer: Preservative: 0.02% Sodium Azide
Constituents: 1% BSA, PBS, pH 7.4
Purity: Immunogen affinity purified
Clonality: Polyclonal
Isotype: IgG

Applications

Our Abpromise guarantee covers the use of ab73400 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 a concentration of 5 µg/ml.</td>
</tr>
</tbody>
</table>
Function
Acts as decoy receptor for RANKL and thereby neutralizes its function in osteoclastogenesis. Inhibits the activation of osteoclasts and promotes osteoclast apoptosis in vitro. Bone homeostasis seems to depend on the local RANKL/OPG ratio. May also play a role in preventing arterial calcification. May act as decoy receptor for TRAIL and protect against apoptosis. TRAIL binding blocks the inhibition of osteoclastogenesis.

Tissue specificity
Highly expressed in adult lung, heart, kidney, liver, spleen, thymus, prostate, ovary, small intestine, thyroid, lymph node, trachea, adrenal gland, testis, and bone marrow. Detected at very low levels in brain, placenta and skeletal muscle. Highly expressed in fetal kidney, liver and lung.

Involvement in disease
Defects in TNFRSF11B are the cause of juvenile Paget disease (JPD) [MIM:239000]; also known as hyperostosis corticalis deformans juvenilis or hereditary hyperphosphatasia or chronic congenital idiopathic hyperphosphatasia. JPD is a rare autosomal recessive osteopathy that presents in infancy or early childhood. The disorder is characterized by rapidly remodeling woven bone, osteopenia, debilitating fractures, and deformities due to a markedly accelerated rate of bone remodeling throughout the skeleton. Approximately 40 cases of JPD have been reported worldwide. Unless it is treated with drugs that block osteoclast-mediated skeletal resorption, the disease can be fatal.

Sequence similarities
Contains 2 death domains. Contains 4 TNFR-Cys repeats.

Post-translational modifications
N-glycosylated. Contains sialic acid residues. The N-terminus is blocked.

Cellular localization
Secreted.

Target

Application | Abreviews | Notes
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IHC-P |  | Use a concentration of 1 - 5 µg/ml. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

WB |  | Use a concentration of 1 µg/ml. Detects a band of approximately 48 kDa (predicted molecular weight: 46 kDa).

Images
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Osteoprotegerin antibody (ab73400)

IHC-P image of Osteoprotegerin staining on Rat kidney sections using ab73400 (1:4000). The sections were deparaffinized and subjected to heat mediated antigen retrieval using citric acid. The sections were blocked using 1% BSA for 10 mins at 21°C. ab73400 was diluted 1:4000 in TBS buffer (containing BSA and Azide) and sections were then incubated with ab73400 for 2 hours at 21°C. The secondary antibody used was Biotin conjugated Goat polyclonal to Rabbit IgG (1:250).

Western blot - Anti-Osteoprotegerin antibody (ab73400)

All lanes: Anti-Osteoprotegerin antibody (ab73400) at 1 µg/ml

Lane 1: SK N BE (Human neuroblastoma) Whole Cell Lysate
Lane 2: K562 (Human erythromyeloblastoid leukemia cell line) Whole Cell Lysate

Lysates/proteins at 10 µg per lane.

Secondary
All lanes: Goat polyclonal to Rabbit IgG - H&L - Pre-Adsorbed (HRP) at 1/3000 dilution

Performed under reducing conditions.

Predicted band size: 46 kDa
Observed band size: 48 kDa
why is the actual band size different from the predicted?
Additional bands at: 105 kDa, 14 kDa. We are unsure as to the identity of these extra bands.

Exposure time: 8 minutes

Osteoprotegerin contains a number of potential glycosylation sites (SwissProt) which may explain its migration at a higher molecular weight than predicted.
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Osteoprotegerin antibody (ab73400)

IHC image of Osteoprotegerin staining in Human Normal Kidney FFPE section, performed on a BondTM system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab73400, 5µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

Immunocytochemistry/ Immunofluorescence - Anti-Osteoprotegerin antibody (ab73400)

ICC/IF image of ab73400 stained MCF7 cells. The cells were 100% methanol fixed (5 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 (ab73400, 5µg/ml) overnight at +4°C. The secondary antibody (green) was Alexa Fluor® 488 goat anti-rabbit IgG (H+L) used at a 1/1000 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 1.43µM. This antibody also gave a positive result in 100% methanol fixed (5 min) HeLa, Hek293 and HepG2 cells at 5µg/ml.

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