Product datasheet

Anti-Osteopontin antibody ab8448

**Overview**

**Product name**
Anti-Osteopontin antibody

**Description**
Rabbit polyclonal to Osteopontin

**Host species**
Rabbit

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

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

**Immunogen**
Synthetic peptide: CKSKKFRRPNQYPD, corresponding to amino acids 170-183 of Human Osteopontin. (Peptide available as ab9078).

**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: 0.1% Sodium azide
Constituents: 0.42% Potassium phosphate, 0.87% Sodium chloride

**Purity**
Whole antiserum

**Clonality**
Polyclonal

**Isotype**
IgG

**Applications**

Our Abpromise guarantee covers the use of ab8448 in the following tested applications.

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

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**Function**

Binds tightly to hydroxyapatite. Appears to form an integral part of the mineralized matrix. Probably important to cell-matrix interaction. Acts as a cytokine involved in enhancing production of interferon-gamma and interleukin-12 and reducing production of interleukin-10 and is essential in the pathway that leads to type I immunity.

**Tissue specificity**

Bone. Found in plasma.

**Sequence similarities**

Belongs to the osteopontin family.

**Post-translational modifications**

Extensively phosphorylated on clustered serine residues. N- and O-glycosylated. Phosphorylation sites are present in the extracellular medium.

**Cellular localization**

Secreted.

**Images**

**All lanes**: Anti-Osteopontin antibody (ab8448) at 1/1000 dilution

**Lane 2**: Human Osteopontin
**Lane 3**: MMP-cleaved Human Osteopontin

Lyssates/proteins at 0.25 µg per lane.

**Secondary**

**All lanes**: HRP-conjugated Goat anti-Rabbit IgG at 1/10000 dilution

The osteopontin antibody (ab8448) is used at 1:1000 dilution on a blot with 250ng human osteopontin (lane 2) and MMP-cleaved osteopontin (lane 3)
ICC/IF image of ab8448 stained MCF7 cells. The cells were 4% PFA 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 (ab8448, 1/1000 dilution) 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.

Immunohistochemistry (Frozen sections) - Anti-Osteopontin antibody (ab8448)

This image is a courtesy of Anonymous Abreview

ab8448 staining Osteopontin in mouse developing skeleton tissue section by Immunohistochemistry (Frozen sections). Tissue samples were fixed with paraformaldehyde before permeabilization with 0.1% Triton and blocking with 20% serum was performed for 1 hour at RT. The sample was incubated with primary antibody (1/200) in 20%FBS/PBS for 16 hours at 4°C. An Alexa Fluor®488-conjugated donkey polyclonal to rabbit IgG was used as secondary antibody at 1/200 dilution. In the image: Red Rhodamine Phalloidin (muscle), Blue DAPI (nuclei), Green Osteopontin.
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Osteopontin antibody (ab8448)

Breast tumour section. Osteopontin is a normal component of elastic fibers in the breast (shown heavily stained in this section). There is also weak staining of the extracellular matrix. Osteopontin is not believed to be expressed inside breast tumour cells, and there is no staining in the intracellular region of the breast cells in this section.

Osteopontin antibody (ab8448) used at 1:100-1:300. No antigen retrieval is required.

ab8448 staining Osteopontin in Mouse femur tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffin-embedded sections). Tissue was fixed with paraformaldehyde and blocked with 5% BSA for 1 hour at 35°C; antigen retrieval was by heat mediation in a citrate buffer. Samples were incubated with primary antibody (1/50) for 14 hours at 4°C. An Alkaline Phosphatase-conjugated Goat anti-rabbit IgG F(ab')2 polyclonal (1/100) was used as the secondary antibody.

OPN is cleaved by MMP to yield 2 fragments, which migrate at 40kD (N terminal) and 32kD (C terminal). The C terminal fragment can undergo further cleavage by both of these MMPs (see Agnihotri et al, JBC 2001 for further details). The epitope recognised by ab8448 is shown in violet. This antibody detects the full length OPN and the 32kD fragment. It does not recognise the 40kD fragment.

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