abcam

Product datasheet

Anti-Lipocalin-2 / NGAL antibody [EPR19912] ab206427





1 References 6 Images

Overview

Product name Anti-Lipocalin-2 / NGAL antibody [EPR19912]

Rabbit monoclonal [EPR19912] to Lipocalin-2 / NGAL **Description**

Host species Rabbit

Tested applications Suitable for: IHC-P, WB

Species reactivity Reacts with: Human

Immunogen Recombinant full length protein. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: SW480 and A431 whole cell lysates; human ovary cancer, fetal spleen and colon cancer

lysates; IHC-P: Human spleen, liver and ovary cancer tissues.

General notes This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility

- Improved sensitivity and specificity

- Long-term security of supply

- Animal-free production

For more information see here.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

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 long

term. Avoid freeze / thaw cycle.

Storage buffer pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

Purity Protein A purified

Clonality Monoclonal Clone number EPR19912

Isotype ΙgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab206427 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
IHC-P		1/8000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
WB		1/1000. Detects a band of approximately 22 kDa (predicted molecular weight: 22 kDa).

Target

Function

Iron-trafficking protein involved in multiple processes such as apoptosis, innate immunity and renal development. Binds iron through association with 2,5-dihydroxybenzoic acid (2,5-DHBA), a siderophore that shares structural similarities with bacterial enterobactin, and delivers or removes iron from the cell, depending on the context. Iron-bound form (holo-24p3) is internalized following binding to the SLC22A17 (24p3R) receptor, leading to release of iron and subsequent increase of intracellular iron concentration. In contrast, association of the iron-free form (apo-24p3) with the SLC22A17 (24p3R) receptor is followed by association with an intracellular siderophore, iron chelation and iron transfer to the extracellular medium, thereby reducing intracellular iron concentration. Involved in apoptosis due to interleukin-3 (IL3) deprivation: iron-loaded form increases intracellular iron concentration without promoting apoptosis, while iron-free form decreases intracellular iron levels, inducing expression of the proapoptotic protein BCL2L11/BIM, resulting in apoptosis. Involved in innate immunity, possibly by sequestrating iron, leading to limit bacterial growth.

Tissue specificity

Expressed in bone marrow and in tissues that are prone to exposure to microorganism. High expression is found in bone marrow as well as in uterus, prostate, salivary gland, stomach, appendix, colon, trachea and lung. Not found in the small intestine or peripheral blood leukocytes.

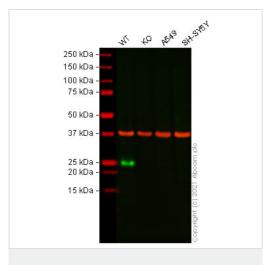
Sequence similarities

Belongs to the calycin superfamily. Lipocalin family.

Cellular localization

Secreted. Upon binding to the SLC22A17 (24p3R) receptor, it is internalized.

Images



Western blot - Anti-Lipocalin-2 / NGAL antibody [EPR19912] (ab206427)

All lanes : Anti-Lipocalin-2 / NGAL antibody [EPR19912] (ab206427) at 1/1000 dilution

Lane 1 : Wild-type SW480 (Human colorectal adenocarcinoma cell line) whole cell lysate

Lane 2: LCN2 knockout SW480 (Human colorectal adenocarcinoma cell line) whole cell lysate

Lane 3: A549 (Human lung carcinoma cell line) whole cell lysate

Lane 4: SH-SY5Y (Human neuroblastoma cell line from bone

marrow) whole cell lysate

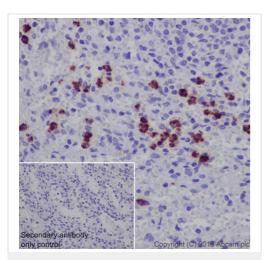
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 22 kDa Observed band size: 25 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab206427 observed at 25 kDa. Red - loading control <u>ab8245</u> (Mouse anti-GAPDH antibody [6C5]) observed at 37 kDa.

ab206427 was shown to react with Lipocalin-2 / NGAL in wild-type SW480 cells in Western blot with loss of signal observed in LCN2 knockout cell line ab270486 (knockout cell lysate ab270509). Wild-type SW480 and LCN2 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with ab206427 and ab8245 (Mouse anti-GAPDH antibody [6C5]) overnight at 4 °C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preabsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Lipocalin-2 / NGAL antibody [EPR19912] (ab206427)

250 kDa -250 kDa -250 kDa -250 kDa -150 kDa -150 kDa -150 kDa -150 kDa -100 kDa -100 kDa -100 kDa -100 kDa -75 kDa -75 kDa -75 kDa -75 kDa -50 kDa -50 kDa -50 kDa -50 kDa -37 kDa -37 kDa -37 kDa -37 kDa -25 kDa -25 kDa -25 kDa 🕳 25 kDa -20 kDa -20 kDa -20 kDa -20 kDa -15 kDa -15 kDa -15 kDa -15 kDa -10 kDa -10 kDa -10 kDa -10 kDa -

Western blot - Anti-Lipocalin-2 / NGAL antibody [EPR19912] (ab206427)

Immunohistochemical analysis of paraffin-embedded human spleen tissue labeling Lipocalin-2 / NGAL with ab206427 at 1/8000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Positive staining on neutrophils is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

All lanes : Anti-Lipocalin-2 / NGAL antibody [EPR19912] (ab206427) at 1/1000 dilution

Lane 1 : A431 (Human epidermoid carcinoma cell line) whole cell lysate

Lane 2 : Human ovary cancer lysate

Lane 3: Human fetal spleen lysate

Lane 4: Human colon cancer lysate

Lysates/proteins at 10 µg per lane.

Secondary

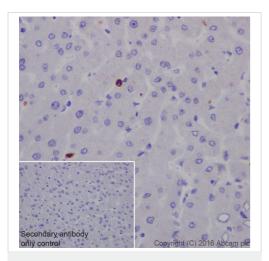
Lane 1: Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000

Lanes 2-4: Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/100000 dilution

Predicted band size: 22 kDa Observed band size: 22 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: Lane 1-3: 3 minutes; Lane 4: 30 seconds.

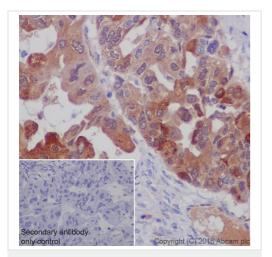


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Lipocalin-2 / NGAL antibody [EPR19912] (ab206427)

Immunohistochemical analysis of paraffin-embedded human liver tissue labeling Lipocalin-2 / NGAL with ab206427 at 1/8000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Positive staining on neutrophils and negative on hepatocytes. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

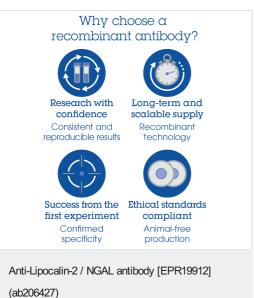


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Lipocalin-2 / NGAL antibody [EPR19912] (ab206427)

Immunohistochemical analysis of paraffin-embedded human ovary cancer tissue labeling Lipocalin-2 / NGAL with ab206427 at 1/8000 dilution, followed by Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution. Cytoplasmic staining on human ovary cancer is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



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