Anti-NOD2, clone 2D9

Monoclonal Antibody Cat. # 04-145 I ot # 2427492

FOR RESEARCH USE ONLY

pack size: 100 µg

Store at 2-8°C



Certificate of Analysis

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Applications	Species Cross-Reactivity	Antibody Isotype	Epitope/ Region	Host Species	Molecular Weight	Accession #
IH, WB, IP	Н	lgG1κ	Unknown	M	110 kDa	NP_071445

Background

NOD2 (nucleotide-binding oligomerization domain containing 2), also known as the caspase recruitment domain family, member 15 (CARD15), is a member of the NOD-like Receptor (NLR) family of proteins and plays an important role in the immune system. It is an intracellular pattern recognition receptor and is linked to inflammatory diseases such as inflammatory bowel disease/Crohn's Disease and Blau syndrome. NOD2 has two caspase recruitment domains (CARDs) and eleven leucine-rich repeats (LRRs). The protein is primarily expressed in the peripheral blood leukocytes. It plays a role in the immune response by recognizing the bacterial molecules. Recently it is has been found to interact with NLRC4 and bind to MAVS in response to ssRNA or viral RNA treatment and activate the IFN response.

Presentation

Purified mouse monoclonal IgG1k in buffer containing 0.1 M Tris-Glycine (pH 7.4, 150 mM NaCl) with 0.05% sodium azide.

Concentration

1.0 mg/mL

Specificity

This antibody recognizes NOD2.

Species Cross-reactivity

Proven to react with human.

Immunogen

His-tagged recombinant protein corresponding to human NOD2.

Molecular Weight

~110 kDa

Method of Purification

Protein G Purified

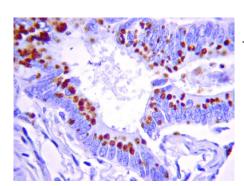
Storage and Handling

Stable for 1 year at 2-8°C from date of receipt.

Hyperplastic colorectal carcinoma tissue

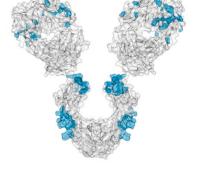
Quality Control Testing

Immunohistochemistry Analysis: dilution of this antibody detected NOD2 in hyperplastic colorectal carcinoma tissue.



Immunohistochemistry Analysis:

Representative lot data. Representative positive and background staining patterns on formalin-fixed, paraffin-embedded hyperplastic colorectal carcinoma. Tissue was pre-treated with citrate pH 6.0 using antigen retrieval techniques. Antibody was diluted to 1:25 and detected using Millipore's IHC Select® HRP/DAB detection kit protocol (Cat .No. DAB050). Immunoreactivity is seen as restricted to metaplastic paneth cells as well as infiltrading LPM cells.



References

- 1. McDonald, C., Lecine, P., et al. (2007). J Biol Chem. 282(20):15197-15207.
- 2. Ogura, Y., et al. (2003). Gut. 52(11):1591-

Additional Research Applications

Western Blot Analysis: A previous lot of this antibody was shown to work in western blot by an independent laboratory (McDonald, C., Lecine, P., et al., 2007).

Immunoprecipitation: A previous lot of this antibody was shown to work immunoprecipitation by an independent laboratory (McDonald, C., Lecine, P., et al., 2007).

PROTOCOL

Immunohistochemistry

- 1. Use standard deparaffinization techniques on tissue specimens.
- Pre-treat tissues using Heat Induced Epitope Retrieval (HIER) techniques (As needed/Primary-antibody-dependent).
 Note: Do not allow tissues to dry out during the staining procedure.

The following steps are taken from the product manual for IHC Select® HRP/DAB Detection Kit (Cat. # DAB050). Utilize either Capillary method or PapPen procedure (PapPen is described below).

- Apply the blocking reagent to the tissue specimen (encircled in a hydrophobic barrier-PapPen) and incubate in a humidifier chamber for 10
 minutes.
- 4. While holding the slide at a 45° angle, gently rinse the specimen with 1X Rinse Buffer for a minimum of 15 seconds. Tap the end of the slide onto a paper towel to remove excess Rinse Buffer.
- Apply a recommended dilution of primary antibody over the entire tissue specimen and incubate in a humidifier chamber at room temperature for 60 minutes (amount of time is primary antibody dependent, as well as titer dependent).
- 6. Rinse specimen as performed in Step 4.
- 7. Apply the biotinylated secondary antibody to the tissue specimen and incubate in a humidifier chamber for 10 minutes.
- 8. Rinse specimen as performed in Step 4.
- 9. Quench endogenous peroxidase by incubating in 3% aqueous hydrogen peroxide for 10 min.
- 10. Apply the Streptavidin-HRP solution to the tissue specimen and incubate in an enclosed chamber for 10 minutes.
- 11. Rinse specimen as performed in Step 4.
- 12. Apply the DAB (chromogen reagent) to the tissue specimen and incubate in a humidifier chamber for 10 minutes.
- 13. Rinse specimen as performed in Step 4.
- 14. Apply the Hematoxylin counterstain solution to the tissue specimen and incubate in an enclosed chamber for 1 minute.
- 15. Rinse specimen as performed in Step 4.
- 16. Place the tissue slides directly into a container filled with de-ionized water until mounting.
- 17. Dehydrate tissue in four changes of 100% reagent alcohol, immerse in 2 changes of xylene, and then apply a xylene-based mounting media (e.g. Permount) and coverslip.

RELATED PRODUCTS (specific) **RELATED PRODUCTS** (non-specific) description cat # description DAB050 IHC Select® HRP/DAB WBAVDBASE SNAP i.d. Protein Detection System WBAVDABTR SNAP i.d. Antibody Collection Tray WBAVDROLL SNAP i.d. Blot Roller WBAVDBH03 SNAP i.d. Triple Well Blot Holder WBAVDBH01 SNAP i.d. Single Well Blot Holder WBAVDBH02 SNAP i.d. Double Well Blot Holder IPVH00010 Immobilon-P 26.5 cm x 3.75 m Roll PVDF 0.45 um IPFL00010 Immobilon-FL 26.5 cm x 3.75 m Roll PVDF 0.45 µm IPVH07850 Immobilon-P 7 x 8.4 cm PVDF 0.45 mm (sheet) 50/pk ISEQ00010 Immobilon-P SQ 26.5 cm x 3.75 m 1 roll PVDF 0.2 μm ISFQ07850 Immobilon-P 7 x 8.4 cm PVDF 0.2 mm (sheet) 50/pk IPFL07810 Immobilon-FL 7 x 8.4 cm PVDF 0.45 mm (sheet) 10/pk WBKLS0100 Immobilon Western Chemilum HRP Substrate 100 mL Spray & Glow™ ECL WB Detection System 1 ea 17-373 Re-Blot Western Blot Recycling Kit 2060 2500 Re-Blot Plus Western Blot Recycling Kit B2080-175GM Blot Quick Blocker Membrane Blocking Agent 175G

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