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AM26352BT-N Monoclonal Antibody to Nitrotyrosine - Biotin

Alternate names: NO-Tyrosine, Nitro-Tyrosine

Quantity: 50 µg

Concentration: 0.1 mg/ml

Background: Nitrotyrosine is formed in tissues in presence of the active metabolite NO and is a stable end product of nitrosylation of tyrosine. Inflammation is characterized by increased nitric oxide (NO) production. NO reacts rapidly with superoxide to form peroxynitrite. At physiological pH and in the presence of transition metals, peroxynitrite undergoes heterolytic cleavage to form hydroxyl anion and nitronium ion, the latter of which nitrates protein tyrosine residues. The presence of nitrotyrosine has been detected in various inflammatory processes including atherosclerotic plaques, Amyotrophic Lateral Sclerosis (ALS) and Multiple Sclerosis (MS). Thus, the presence of nitrotyrosine on proteins can be used as a marker for peroxynitrite formation in vivo and consequently as a marker of NO-mediated tissue damage.

Host / Isotype: Mouse / IgG2b

Recommended Isotype Controls: SM12B

Clone: HM.11

Immunogen: Nitrated KLH

Format:
State: Liquid 0.2 µm filtered Ig fraction
Purification: Protein G
Buffer System: PBS
Stabilizers: 0.1% bovine serum albumin
Label: Biotin

Applications: Immunohistochemistry on frozen sections (1,3): Also cytospins: acetone fixation 10 min -20 °C; block endogenous peroxidase by 0.3 % H2O2 in PBS (or methanol for intracellular staining); blocking with 10% NGS or 5 % BSA for 30 min. The typical starting working dilution is 1:10.
Immunohistochemistry on paraffin sections (4,5): 10% formalin fixation; 3% H2O2 to block endogenous peroxidases; Citrate buffer pH 6.0 for 1 min at 100 °C as antigen retrieval treatment. Positive control: Human lung tissue. The typical starting working dilution is 1:10.
Immunoassays (1).
Western blot (2-5): Reduced and no-reduced samples; block with 5% BSA or skimmed milk. Positive control: mouse kidney lysate, mouse optic nerve, retina , spinal cord and brain lysates, rat aorta lysate. The typical starting working dilution is 1:10.
Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.

Specificity:

The monoclonal antibody HM.11 recognizes modified amino acid nitrotyrosine in all different species. It recognizes nitrotyrosine, both with the free amino acid as well as with proteins containing nitrotyrosine.
Does not cross react with Phosphotyrosine or Chlorotyrosine.

Storage:

Store at 2 - 8 °C.
Shelf life: one year from despatch.

Caution:

Keep sterile, do not add sodium azide or any nitric containing preservative since this will affect the antibody!

General Readings:

1. ter Steege JC, Koster-Kamphuis L, van Straaten EA, Forget PP, Buurman WA. Nitrotyrosine in plasma of celiac disease patients as detected by a new sandwich ELISA. Free Radic Biol Med. 1998 Nov 15;25(8):953-63. PubMed PMID: 9840741.
2. Beckmann JS, Ye YZ, Anderson PG, Chen J, Accavitti MA, Tarpey MM, et al. Extensive nitration of protein tyrosines in human atherosclerosis detected by immunohistochemistry. Biol Chem Hoppe Seyler. 1994 Feb;375(2):81-8. PubMed PMID: 8192861.
3. Kooy NW, Royall JA, Ye YZ, Kelly DR, Beckman JS. Evidence for in vivo peroxynitrite production in human acute lung injury. Am J Respir Crit Care Med. 1995 Apr;151(4):1250-4. PubMed PMID: 7697261.
4. Beckman JS, Beckman TW, Chen J, Marshall PA, Freeman BA. Apparent hydroxyl radical production by peroxynitrite: implications for endothelial injury from nitric oxide and superoxide. Proc Natl Acad Sci U S A. 1990 Feb;87(4):1620-4. PubMed PMID: 2154753.

Pictures:

Nitrotyrosine in human liver of severely obesed patients. Staining of paraffin tissue section with clone HM.11 (Cat. # AM26352PU-N). Anti-nitrotyrosine at 2µg/ml (1h, RT)