

PRODUCT INFORMATION



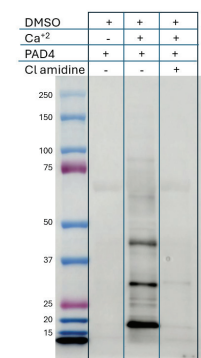
Anti-Citrulline Monoclonal Antibody (Clone 1D9)

Item No. 30773

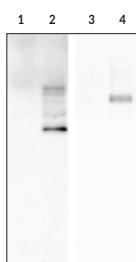
Overview and Properties

Contents:	This vial contains 100 µg of protein G-purified monoclonal antibody.
Synonyms:	Citrullinated Protein, Pan-citrulline, Peptidyl-citrulline
Immunogen:	Citrulline-containing peptide conjugated to keyhole limpet hemocyanin
Cross Reactivity:	(+) Citrullinated proteins; (-) Native proteins
Species Reactivity:	Species Independent
Form:	Liquid
Storage:	-20°C (as supplied)
Stability:	≥3 years
Storage Buffer:	PBS, pH 7.2, with 50% glycerol and 0.02% sodium azide
Clone:	1D9
Host:	Mouse
Isotype:	IgG2b
Applications:	Western Blot (WB), ELISA, and Immunoprecipitation (IP); the recommended starting dilution for WB is 1:1000 and 1:200 for ELISA and IHC. The recommended concentration for IP is 5-10 µg of antibody per test. Other applications were not tested, therefore optimal working concentration/dilution should be determined empirically.

Images



LS 1747 cells were treated with the indicated conditions. Cells were lysed, and tested by WB using Cayman's Anti-Citrulline Monoclonal Antibody (Clone 1D9) (Item No. 30773) at 1:1,000 and Cayman's Goat Anti-Mouse IgG HRP secondary (Item No. 10004302) at 1:10,000.

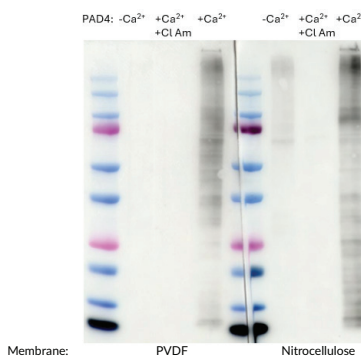


Lanes 1 and 2 were immunoprecipitated using the Anti-Citrulline Monoclonal Antibody (Clone 1D9) (Item No. 30773) and probed with a GFAP Polyclonal Antibody (Item No. 28848).

Lane 1: GFAP (human, recombinant) (Item No. 27353)
Lane 2: Citrullinated GFAP (human, recombinant) (Item No. 28622)

Lanes 3 and 4 were immunoprecipitated using the Anti-Citrulline Monoclonal Antibody (Clone 1D9) (Item No. 30773) and probed with a Fibrinogen (α chain) Polyclonal Antibody (Item No. 18033).

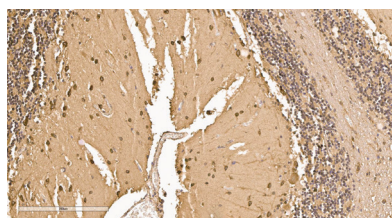
Lane 3: Human Fibrinogen
Lane 4: Human Fibrinogen (PAD4 Citrullinated) (Item No. 400076)



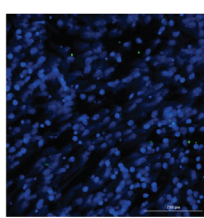
Membrane:

Primary Ab: IgG clone ID9 30773 used at 1 µg/ml, for 1 hour, rt in 2% BSA/PBS, then PBS washes
Secondary Ab: 10004302 Goat anti-mouse IgG HRP (1:10,000) in 2% BSA/PBS, then PBS washes
Imaging: 25 seconds capture with Pico ECL substrate (Thermo)

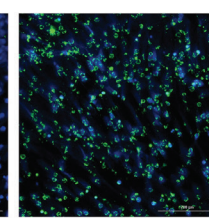
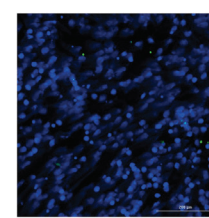
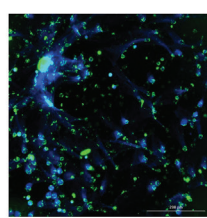
PAD4 (human, recombinant) (Item No. 10500) treated U937 lysate (20 µg/lane) with or without calcium or Cl-amidine.



Immunohistochemistry analysis of formalin-fixed, paraffin-embedded (FFPE) human Alzheimer's brain, cerebellum, tissue after heat induced antigen retrieval in pH 6.0 citrate buffer. After incubation with Anti-Citrulline Monoclonal Antibody (Clone 1D9) (Item No. 30773) at a 1:200 dilution, slides were incubated with biotinylated secondary antibody, followed by alkaline phosphatase-streptavidin and chromogen (DAB).



Primary neutrophils induced with 100 nM PMA to produce neutrophil extracellular traps (NETs) were fixed with 3.7% PFA and blocked with 5% normal goat serum. Cells were probed with a mouse IgG1 isotype control (A) or the Anti-Citrulline Monoclonal Antibody (Clone 1D9) (Item No. 30773) at a 1:200 dilution (B) followed by secondary antibody Goat Anti-Mouse (IgG+IgM) FITC (Item No. 10006617). Cell nuclei were stained with DAPI.



Primary neutrophils induced with 100 nM PMA to produce neutrophil extracellular traps (NETs) were fixed with 3.7% PFA and blocked with 5% normal goat serum. Cells were probed with a mouse IgG1 isotype control (A) or the Anti-Citrulline Monoclonal Antibody (Clone 1D9) (Item No. 30773) at a 1:200 dilution (B) followed by secondary antibody Goat Anti-Mouse (IgG+IgM) FITC (Item No. 10006617). Cell nuclei were stained with DAPI.

WARNING
THIS PRODUCT IS FOR RESEARCH ONLY - NOT FOR HUMAN OR VETERINARY DIAGNOSTIC OR THERAPEUTIC USE.

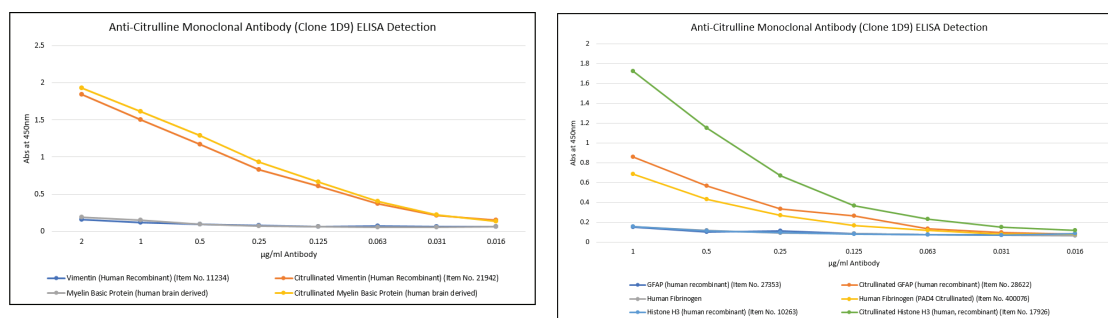
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PRODUCT INFORMATION



Description

Citrulline is a noncoding amino acid that is produced by deimination of arginine through the post-translational modification citrullination.¹ Citrullination is catalyzed by protein arginine deiminases (PADs) that convert positively charged arginine to electrically neutral citrulline, decreasing the isoelectric point of the protein, altering the native protein structure, and influencing protein ionic interactions.² Protein citrullination has roles in many physiological and pathological processes, including autoimmunity, cancer, and neurodegenerative disorders.³ Citrullination of arginine 26 on histone H3 by PAD2 (Item No. 10785) displaces histone H3 from chromatin, resulting in chromatin decondensation and estrogen receptor α (ER α) transcriptional activation in a reporter assay.⁴ Citrullinated histones are also a component of neutrophil extracellular traps (NETs), a network of decondensed DNA and intracellular proteins secreted by neutrophils as a pathogen defense mechanism that is also a source of citrullinated autoantigens.⁵ Increased levels of antibodies to citrullinated protein antigens (ACPAs) are associated with increased disease severity in patients with rheumatoid arthritis.^{6,7} Plasma levels of citrullinated histone H3 are increased in patients with advanced cancer, and citrullinated glial fibrillary acidic protein (citGFAP; Item No. 28622) has been found in postmortem hippocampus from patients with Alzheimer's disease.^{8,9} Cayman's Anti-Citrulline Monoclonal Antibody (Clone 1D9) can be used for ELISA, immunohistochemistry (IHC), and immunoprecipitation (IP) applications.

References

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