Director/Scientist : Jean.Sevigny@crchul.ulaval.ca
Production of Abs, sales and shipping : Mrs Julie Pelletier, info@ectonucleotidases-ab.com
Web site: www.ectonucleotidases-ab.com

Size	Price
25 µl of serum	Contact us

Polyclonal guinea pig anti-mouse NTPDase3 antibodies

Name: mN3-1$_C$I$_4$; mN3-2$_C$(I$_4$,I$_5$); mN3-3$_C$(I$_4$,I$_5$)

Applications[1]

	Yes	Dilution	No	Not tested
Western blot (non-reduced)§	+	1:500-1:2000		
Western blot (reduced)¶	+	1:500		
Immunohistochemistry*	+	1:1000		
Flow cytometry	+	1:100-1:250		
ELISA				×
Immunoprecipitation				×

§ Due to the low abundance of NTPDase3 in tissues and/or due to a weak reactivity of the antibody in Western blot these antibodies give a hardly visible signal on tissues homogenate. A partial purification of the antigen as CONA column is necessary.

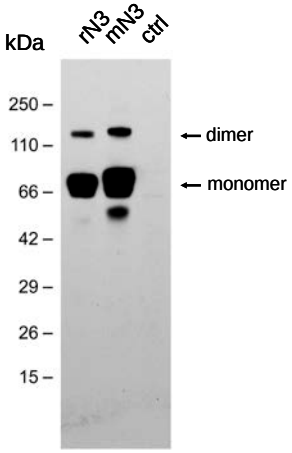
¶ Only mN3-1$_C$I$_4$ work in reduced condition, but this antibody work better in non-reduced conditions (i.e. in absence of DTT, mercaptoethanol), all the other antibodies do not work in reduced conditions.

* Cryosection and acetone fixation

Cross-reactivity

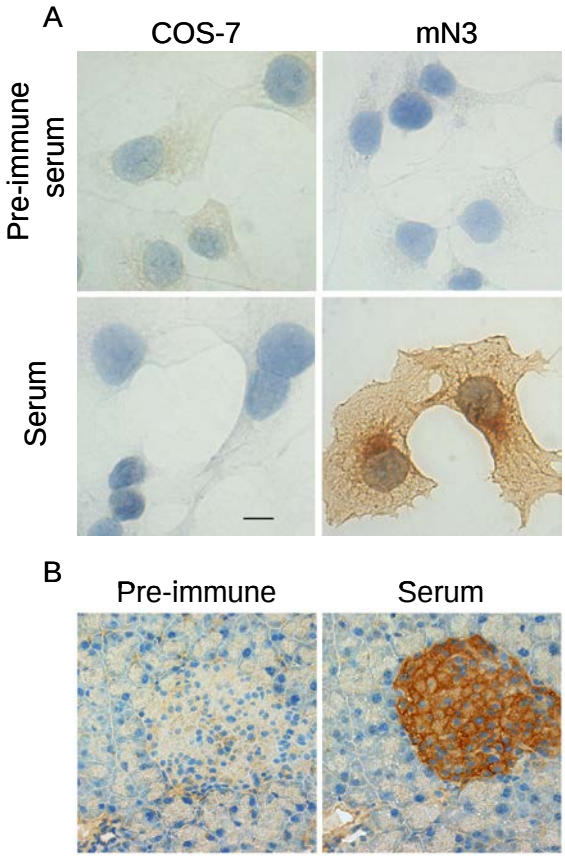
In Western blot and Immunohistochemistry, mN3-1$_C$I$_4$ cross-reacts with rat NTPDase3 but not with mouse NTPDase1-2 and 8.

Western blot[1]



Protein samples (6 µg) from a lysate from HEK 293 cells (ctrl), transiently transfected with a plasmid encoding for rat (r) or mouse (m) NTPDase3 were loaded on NuPAGE® Novex® Bis-Tris 4-12% gels under non-reducing conditions, transferred to an Immobilon-P membrane and incubated with mN3-1$_C$I$_4$ (1:1000). A specific band is detected in the protein samples from cells expressing mouse or rat NTPDase3 at the right molecular weights. Note that NTPDase3 is generally seen as both a monomer and a dimer in Western blot performed in these conditions.

Immuno(cyto/histo)chemistry[1]

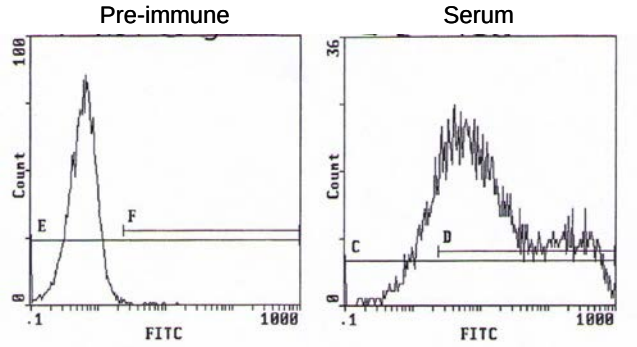


A: Immunocytochemistry of untransfected COS-7 cells (COS-7) or transfected with a plasmid encoding mouse NTPDase3 (mN3) both incubated with either mN3-$1_C I_4$ or preimmune serum. A strong signal is observed only with the antiserum in cells expressing mouse NTPDase3. No signal is detected in any of the controls cells.

B: A mouse pancreas section incubated with either mN3-$1_C I_4$ or preimmune serum. The antiserum displays a positive reaction on the cells of the Langerhans ilets.

In both panels, nuclei are stained with hematoxylin (blue).

Flow cytometry[1]



HEK 293 cells transfected with mouse NTPDase3 cDNA vector. Cells were incubated with mN3-$1_C I_4$ (1:100) or preimmune serum at the same dilution. Transfected cells incubated with the antiserum show a rightward shift.

Storage

To avoid excessive freeze-thaw cycles, a small amount can be kept at 4°C for generally up to one year. A better method consists to dilute the antibody 10 times in one part of 145 mM NaCl, 1% BSA, 10 mM Tris (pH 7.4), and one part of glycerol (for a final concentration of 50% v/v) and to keep it at -20°C (note that 50% glycerol solutions freeze at about -30°C). For long-term storage, freeze samples directly at -80°C.

1:Protocols and experimental conditions are available at http://www.ectonucleotidases-ab.com/Protocols.php.

Reference to cite in your publication (paper where these antibodies were characterized)

This antibody was obtained from ectonucleotidases-ab.com and its specificity was characterized in:
Martín-Satué M, Lavoie EG, Pelletier J, Fausther M, Csizmadia E, Guckelberger O, Robson SC, Sévigny J. Localization of plasma membrane bound NTPDases in the murine reproductive tract. *Histochem Cell Biol.* 2009; 131(5):615-628.

Few other references where these antibodies were used

- Martín-Satué M, Lavoie EG, Fausther M, Lecka J, Aliagas E, Kukulski F, Sévigny J. High expression and activity of ecto-5'-nucleotidase/CD73 in the male murine reproductive tract. *Histochem Cell Biol.* 2010; 133(6):659-668.
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- Zanin RF, Braganhol E, Bergamin LS, Campesato LF, Filho AZ, Moreira JC, Morrone FB, Sévigny J, Schetinger MR, de Souza Wyse AT, Battastini AM. Differential macrophage activation alters the expression profile of NTPDase and ecto-5'-nucleotidase. *PLoS One*. 2012; 7(2):e31205.
- Zanin RF, Bergamin LS, Braganhol E, Sévigny J, de Souza Wyse AT, Battastini AM. Homocysteine modifies extracellular ATP availability in macrophages. *Toxicol In Vitro*. 2013; 27(8):2273-2278.
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1: Protocols and experimental conditions are available at http://www.ectonucleotidases-ab.com/Protocols.php.