

# Fluorescently Labeled Antibodies

## For Direct Immunohistochemical & Immunocytochemical Applications



Alomone Labs is pleased to offer a new line of its well-characterized antibodies directly conjugated to a new generation of specially developed, bright fluorescent dyes by ATTO-TEC.

ATTO dyes are well known in the field of fluorescent technology and are characterized by strong absorption (high extinction coefficient), high fluorescence quantum yield, and high photo-stability. ATTO dyes are analogous to the established Alexa dyes and were found to be comparable to any fluorescent technology in the market.

The proven quality of the Alomone antibodies together with the bright ATTO dyes generate an invaluable tool that undergoes careful quality control and is specially suited for applications that require simultaneous labeling of different markers.

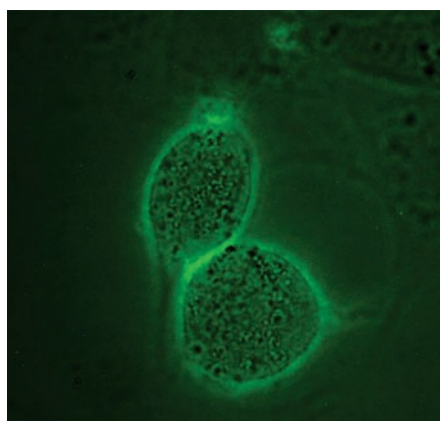
Alomone Labs is currently offering primary antibodies directly conjugated to the following ATTO dyes: ATTO-488 (green) and ATTO-550 (orange).

Label	Fluorescence	Spectra	Alternative to
ATTO-488	Green	$\lambda_{ex}$ 498 nm; $\lambda_{em}$ 520 nm in 0.1 M phosphate pH 7.0	FITC, Alexa-488
ATTO-550	Orange	$\lambda_{ex}$ 554 nm; $\lambda_{em}$ 576 nm in 0.1 M phosphate pH 7.0	TAMRA, Cy3, Alexa-555

### Anti-Angiotensin II Receptor Type-2 (AT<sub>2</sub> Receptor) (extracellular)-ATTO-488

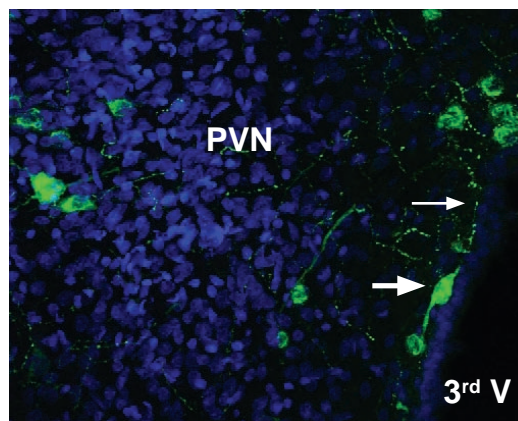
Product#: AAR-012-AG

#### Expression of AT<sub>2</sub> Receptor in mouse 3T3-L1 cells



Immunocytochemical staining of intact live mouse 3T3-L1 cells with Anti-AT<sub>2</sub> Receptor (extracellular)-ATTO-488 antibody (#AAR-012-AG) (green) (1:50). Live view of the same field was superimposed to the fluorescent one.

#### Expression of AT<sub>2</sub> Receptor in rat brain

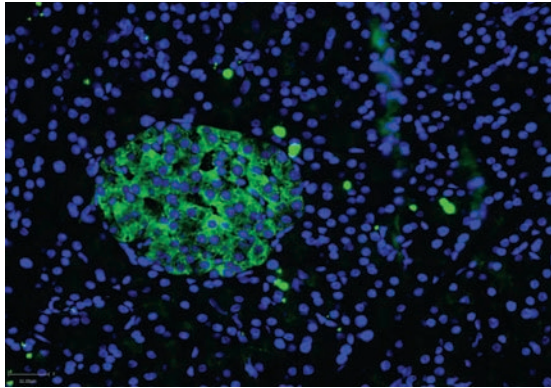


Immunohistochemical staining of AT<sub>2</sub> receptor in rat brain. Rat frozen free floating sections were stained with Anti-AT<sub>2</sub> Receptor (extracellular)-ATTO-488 antibody (#AAR-012-AG) (1:20). The AT<sub>2</sub> receptor (green) was detected in neurons in the vicinity of the hypothalamic paraventricular nucleus (PVN). In some neurons (thick white arrow), axonal processes with varicosities were observed (thin white arrow). Nuclei were visualized with DAPI counterstain (blue).

## Anti-Ca<sub>v</sub>1.2-ATTO-488

Product#: ACC-003-AG

### Expression of Ca<sub>v</sub>1.2 channel in rat pancreas



Immunohistochemical staining of paraffin embedded section of rat pancreas using Anti-Ca<sub>v</sub>1.2-ATTO-488 antibody (#ACC-003-AG) (1:50) (green). Staining is highly specific for endocrine cells of the Islet of Langerhans (IL).

### Expression of Ca<sub>v</sub>1.2 channel in mouse pancreatic microvascular endothelial cells (MS1)

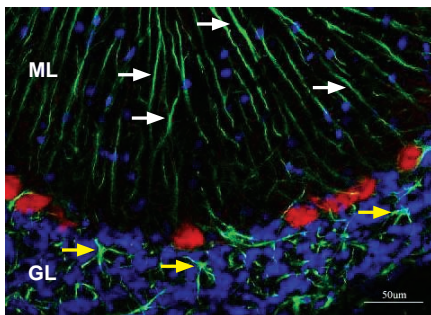


Immunocytochemical staining of MS1 cells using Anti-Ca<sub>v</sub>1.2-ATTO-488 antibody (#ACC-003-AG) (green) (1:50). A, Intracellular staining of Paraformaldehyde-fixed and permeabilized MS1 cells. B, The cell-permeable dye Hoechst 33342 (blue) was used for nuclear staining. C, Merged images of panels A and B.

## Anti-K<sub>ir</sub>4.1-ATTO-488

Product #: APC-035-AG

### Immunohistochemical staining of K<sub>ir</sub>4.1 in rat cerebellum

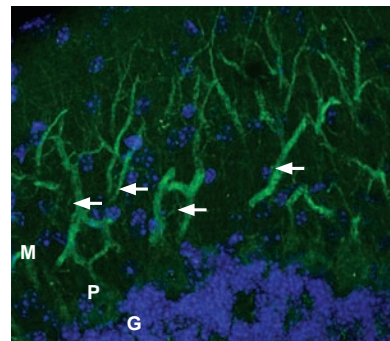


Immunohistochemical staining of frozen sections of rat cerebellum using Anti-K<sub>ir</sub>4.1-ATTO-488 antibody (#APC-035-AG) (green) (1:50). Staining is specific for Bergmann glial cells prolongations (white arrows) in the molecular layer (ML) and astrocytes (yellow arrows) in the granular layer (GL). Purkinje cell bodies are stained with fluorescent Nissl stain (red). Hoechst 33342 (blue) is used as counterstain.

## Anti-AMPA Receptor 2 (GluR2) (extracellular)-ATTO-488

Product #: AGC-005-AG

### Expression of GluR2 in mouse cerebellum

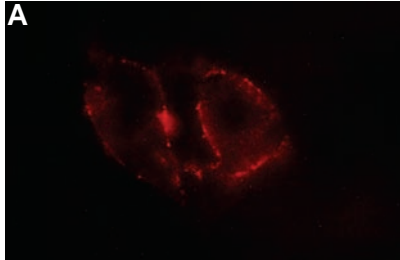


Distribution of GluR2 in the molecular layer of the mouse cerebellum. Frozen free-floating sections were stained with Anti-AMPA Receptor 2 (GluR2) (extracellular)-ATTO-488 antibody (#AGC-005-AG) (green) (1:20). Both dendrites of Purkinje cells (horizontal arrows) were stained (green). DAPI counterstain (blue) helps define the layers: granule (G), Purkinje (P), and molecular (M).

## Anti-Angiotensin II Receptor Type-1 (AT<sub>1</sub> Receptor) (extracellular)-ATTO-550

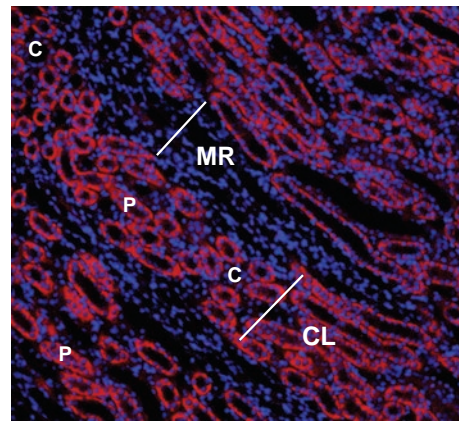
Product#: AAR-011-AO

### Expression of AT<sub>1</sub> Receptor in rat C6 glioma cells



Immunocytochemical staining of live intact rat C6 glioma cells with Anti-AT<sub>1</sub> receptor (extracellular)-ATTO-550 antibody (#AAR-011-AO) (A). live view of the same field (B).

### Expression of AT<sub>1</sub> Receptor in rat kidney

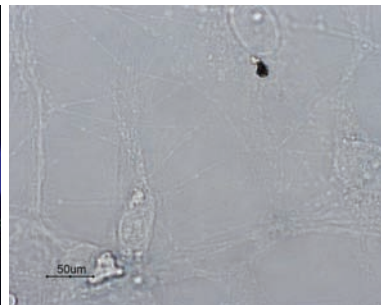
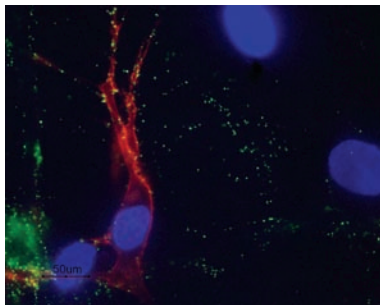


Immunohistochemical staining of AT<sub>1</sub> receptor in rat kidney. Paraffin embedded section of rat kidney showing the most inner layer of the cortex. Notice that intense stain is present in proximal tubules (P) but not in collecting ducts (C) in the cortical labyrinths (CL). Also note that no staining is present both in thin portions of the Loop of Henle or in the collecting ducts in the medullary rays (MR). Slides were treated with citrate for antigen retrieval and then incubated overnight at 4°C with Anti-AT<sub>1</sub> Receptor (extracellular)-ATTO-550 antibody (#AAR-011-AO) (1:50) (Red). Nuclei are visualized with Hoechst 33342 (blue).

## Anti-Human p75<sup>NTR</sup> (extracellular)-ATTO-550

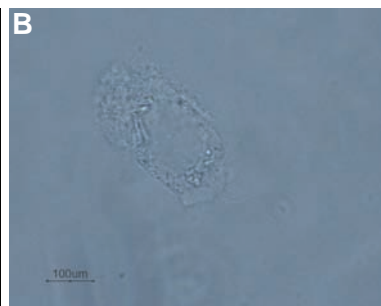
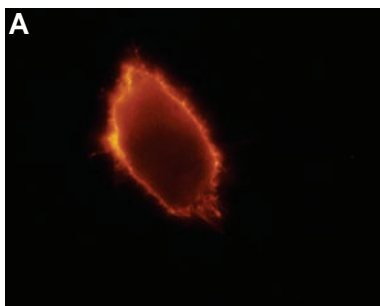
Product#: ANT-007-AO

### Expression of p75<sup>NTR</sup> in rat dorsal root ganglion (DRG) cells



Surface expression of p75<sup>NTR</sup> in rat dorsal root ganglion (DRG) cells. Intact live DRG neurons were labeled with Anti-mGluR1 (extracellular) antibody (#AGC-006) (1:100) followed by goat-anti-rabbit Alexa 488 secondary antibody (green). The cells were then labeled with Anti-Human p75<sup>NTR</sup> (extracellular) ATTO-550 antibody (#ANT-007-AO) (1:50) (red). Nuclei were visualized using the cell-permeable DNA binding dye Hoechst 33342 (blue). Note that the Anti-Human p75<sup>NTR</sup> (extracellular) ATTO-550 antibody does not stain all DRG neurons as expected. Colocalization is observed in some nerve fibers of the mGluR1 and p75<sup>NTR</sup> receptors.

### Expression of p75<sup>NTR</sup> in p75<sup>NTR</sup>-transfected 3T3 cells

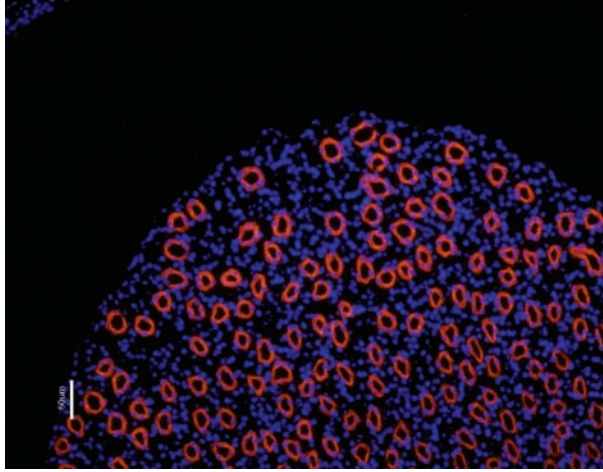


Immunocytochemical staining of p75<sup>NTR</sup> in p75<sup>NTR</sup>-transfected 3T3 cells. (A) Staining of intact live p75<sup>NTR</sup>-transfected 3T3 cells with Anti-Human p75<sup>NTR</sup> (extracellular)-ATTO-550 antibody (#ANT-007-AO) (1:100). (B) Live view of the same field as in (A).

## Anti-Aquaporin 2-ATTO-550

Product#: *AQP-002-AO*

### Expression of Aquaporin 2 in rat kidney

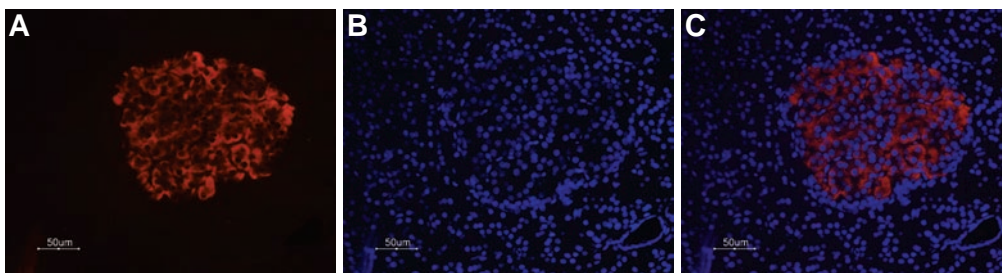


Immunohistochemical staining of paraffin embedded region of rat kidney showing a transversal cut of the inner medulla near to the renal papilla. Aquaporin 2 is detected in collecting ducts but not in thin segments of the loop of Henle. Slides were treated with citrate for antigen retrieval and then incubated overnight at 4°C with **Anti-Aquaporin 2-ATTO-550** antibody (#AQP-002-AO) (1:50) (Red). Nuclei are visualized with Hoechst 33342 (blue).

## Anti-STIM1 (extracellular)-ATTO-550

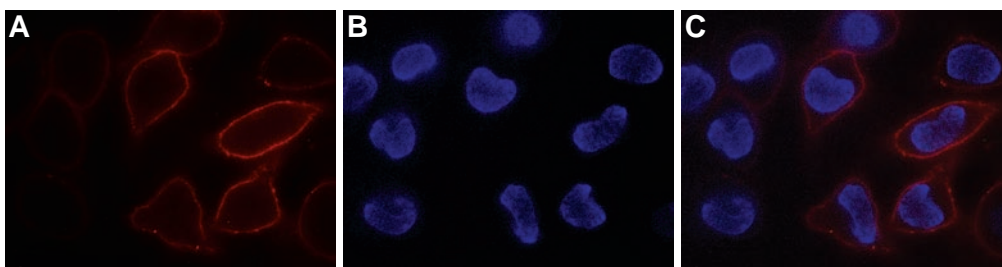
Product#: *ACC-063-AO*

### Expression of STIM1 in rat pancreas



A. Immunohistochemical staining of STIM1 in rat paraffin-embedded pancreas sections using **Anti-STIM1 (extracellular)-ATTO-550** antibody (#ACC-063-AO) (1:20) (red). Staining is highly specific for endocrine cells in the Isle of Langerhans.  
B. Hoechst 33342 (blue) is used as the counterstain.  
C. Merged images of panels A and B.

### Expression of STIM1 in rat basophilic leukemia cells (RBL)

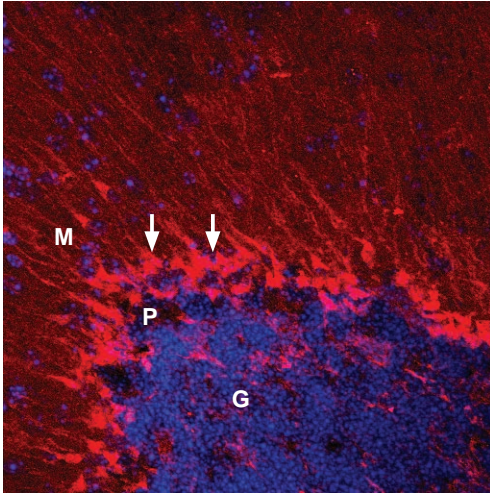


Immunocytochemical staining of STIM1 in live RBL cells  
A. Extracellular staining of cells with **Anti-STIM1 (extracellular)-ATTO-550** antibody (#ACC-063-AO) (1:20) (red).  
B. Nuclear staining of cells using the cell-permeable dye Hoechst 33342.  
C. Merged images of panels A and B.

## Anti-K<sub>v</sub>1.5-ATTO-550

Product #: APC-004-AO

### K<sub>v</sub>1.5 expression in mouse cerebellum

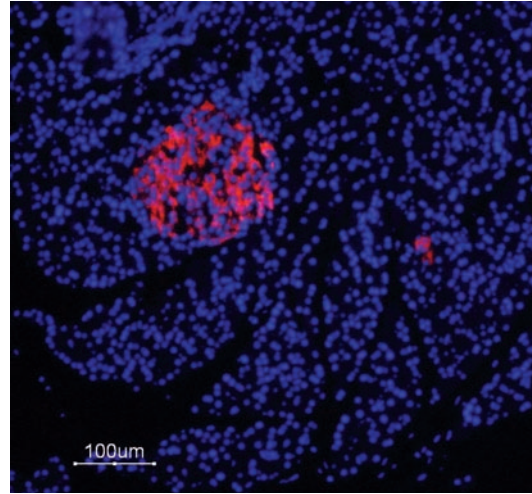


Perfusion fixed, frozen free-floating mouse brain sections were stained with Anti-K<sub>v</sub>1.5-ATTO-550 (#APC-004-AO) (1:50) (red). Staining was detected in cerebellar Bergmann glial cells (white arrows). The blue (DAPI) is a counterstain visualizing nuclei of all cells. G = granule layer, P = Purkinje layer, M = molecular layer.

## Anti-P2X7-ATTO-550

Product#: APR-004-AO

### Expression of P2X7 in rat pancreas

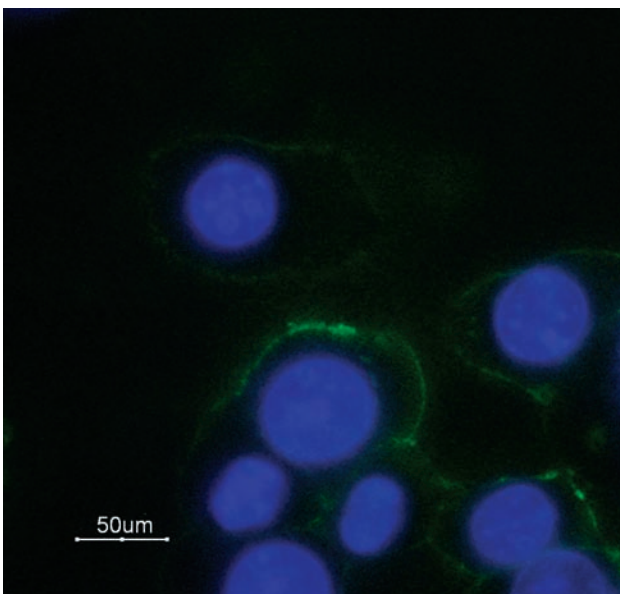


Immunohistochemical staining of P2X7 in rat paraffin embedded endocrine and exocrine pancreas sections using Anti-P2X7-ATTO-550 antibody (#APR-004-AO) (1:20) (red). Staining is highly specific for endocrine cells of the Isle of Langerhans. Counterstain is Hoechst 33342 (blue).

## Anti- $\alpha_{1B}$ -Adrenoceptor-ATTO-488 (extracellular)

Product #: AAR-018-AG

### Immunocytochemical staining of $\alpha_{1B}$ -Adrenoceptor in living GH3 cells



A. Extracellular staining of cells with Anti- $\alpha_{1B}$ -Adrenoceptor-ATTO-488 (extracellular) antibody (#AAR-018-AG) (1:100) (green). Nuclear staining of cells using the cell-permeable dye Hoechst 33342 (blue).

# Fluorescently Labeled Antibodies

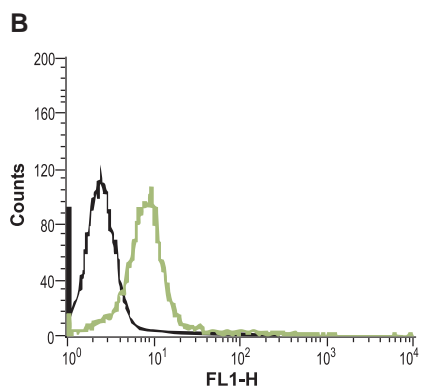
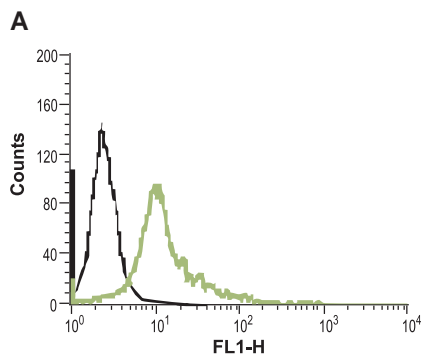
## For Direct Flow Cytometry Analysis



Flow cytometry is a technique for counting, examining, and sorting microscopic particles suspended in a stream of fluid. It allows simultaneous multiparametric analysis of the physical and/or chemical characteristics of single cells flowing through an optical and/or electronic detection apparatus. The flow cytometer was developed in the 1970's and rapidly became an essential instrument for biological studies. Extracellular antibodies directly labeled with fluorescein are an important tool for this application.

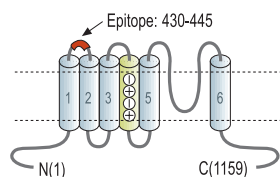
### Anti-K<sub>v</sub>11.1 (HERG) (extracellular)-FITC

Product#: APC-109-F



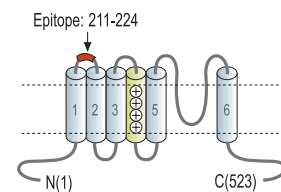
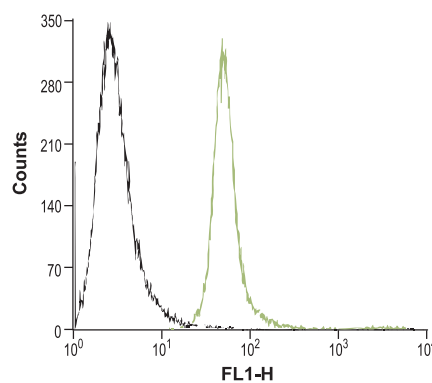
Flow cytometry analysis of intact living K562 (human chronic myelogenous leukemia) cells (A) and Jurkat (human T cell leukemia) cells (B):

— Unstained cells  
 — Cells + Anti- K<sub>v</sub>11.1-(extracellular)-FITC antibody (#APC-109-F)



### Anti-K<sub>v</sub>1.3 (extracellular)-FITC

Product#: APC-101-F

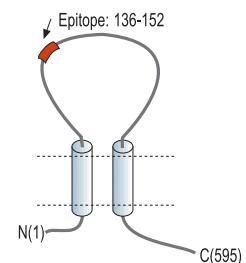
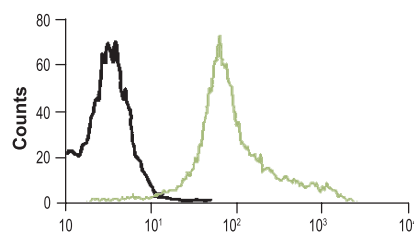


Flow cytometry analysis of intact living Jurkat T-cells:

— Unstained cells.  
 — Cells + anti-K<sub>v</sub>1.3 (extracellular)-FITC antibody (#APC-101-F).

### Anti-P2X7 (extracellular)-FITC

Product#: APR-008-F

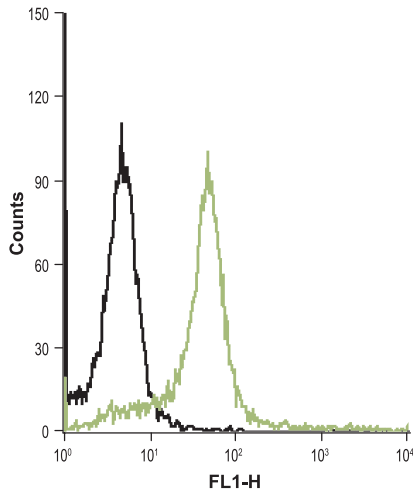


Flow cytometry analysis of intact living Jurkat T-cells:

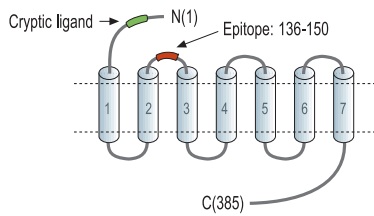
— Unstained cells.  
 — Cells + Anti-P2X7 (extracellular)-FITC antibody (#APR-008-F).

## Anti-Proteinase-Activated Receptor-4 (extracellular)-FITC

Product#: APR-034-F

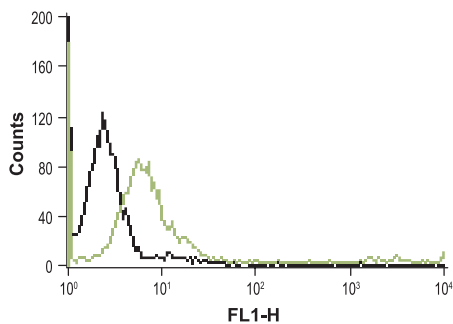


Flow cytometry analysis of PAR-4 expression in live intact HL-60 (human promyelocytic leukemia) cell line:  
 — Unstained HL-60 cells.  
 — HL-60 cells + Anti-Proteinase-Activated Receptor-4 (extracellular)-FITC antibody (#APR-034-F).

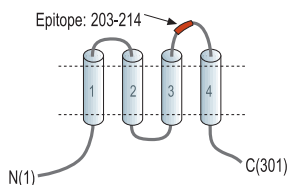


## Anti-Human Orai1 (extracellular)-FITC

Product#: ACC-060-F



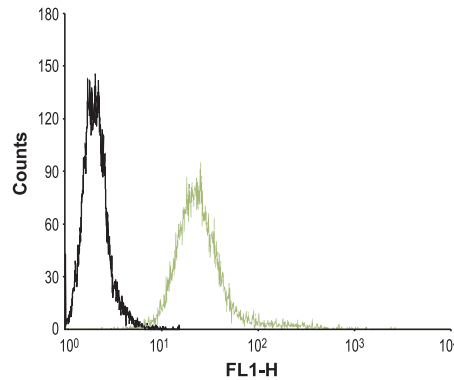
Flow cytometry analysis of live Jurkat (human T cell leukemia) cells:  
 — Unstained cells.  
 — Cells + Anti-Human Orai1 (extracellular)-FITC antibody (#ACC-060-F), (10 µg per 1x10<sup>6</sup> cells).



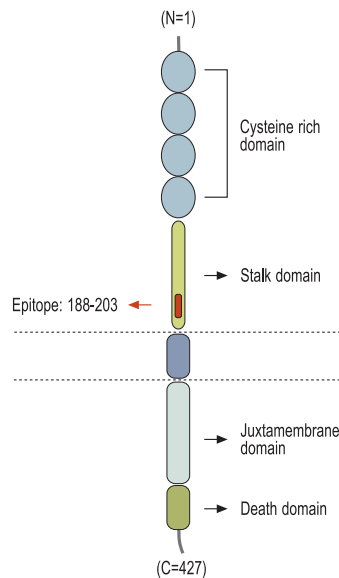
## Anti-Human p75<sup>NTR</sup>-FITC

Product#: ANT-007-F

Host: Rabbit (Polyclonal).  
 Reactivity Confirmed: R, H



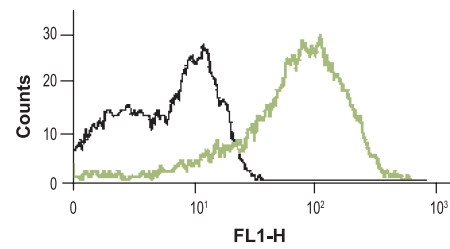
Flow cytometry analysis of live intact rat glioma C6 cells:  
 — Unstained cells.  
 — Cells + Anti-Human p75<sup>NTR</sup>-FITC antibody (#ANT-007-F).



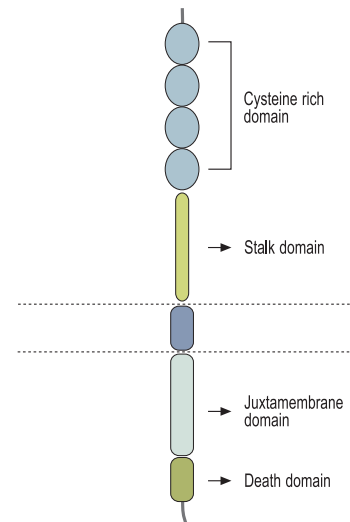
## Anti-Rat p75<sup>NTR</sup>-FITC

Product#: AN-170-F

Host: Mouse (Monoclonal).  
 Reactivity Confirmed: Rat only

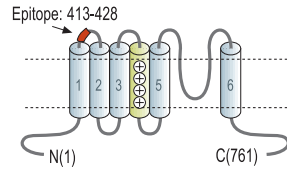
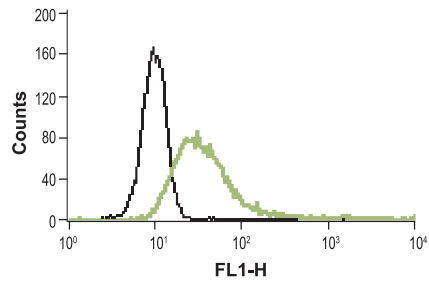


Flow cytometry analysis of PC12 cells:  
 1 µl per 1x10<sup>6</sup> cells (1:50 dilution).  
 — Unstained cells.  
 — Cells + Anti-Rat p75<sup>NTR</sup>-FITC antibody (#AN-170-F).



## Anti-TRPV2 (extracellular)-FITC

Product#: ACC-039-F

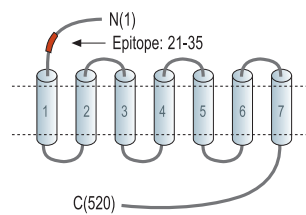
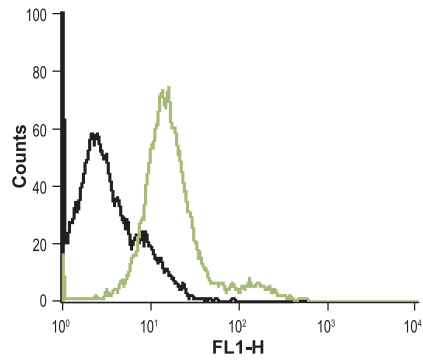


Flow cytometry analysis of intact living RBL cells:

- Unstained cells.
- Cells + Anti-TRPV2 (extracellular)-FITC antibody, (#ACC-039-F).

## Anti- $\alpha_{1B}$ -Adrenoceptor-ATTO-488 (extracellular)

Product #: AAR-018-AG



Flow cytometry analysis of intact living GH3 cells:

- Unstained cells.
- Cells + Anti- $\alpha_{1B}$ -Adrenoceptor-ATTO-488 (extracellular) antibody (#AAR-018-AG) (10 $\mu$ g/5x10<sup>5</sup> cells).