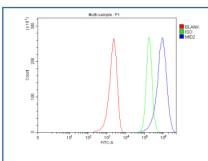


# MID2 Antibody / Midline 2 (FY12728)

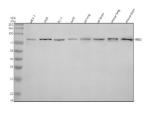
Catalog No.	Formulation	Size
FY12728	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml	100 ug

### **Bulk quote request**

Availability	1-2 days
Species Reactivity	Human, Mouse, Rat
Format	Lyophilized
Clonality	Polyclonal (rabbit origin)
Isotype	Rabbit IgG
Purity	Immunogen affinity purified
Buffer	Each vial contains 4 mg Trehalose, 0.9 mg NaCl, 0.2 mg Na2HPO4.
UniProt	Q9UJV3
Applications	Western Blot : 0.25-0.5ug/ml Flow Cytometry : 1-3ug/million cells
Limitations	This MID2 antibody is available for research use only.



Flow Cytometry analysis of MCF-7 cells using anti-MID2 antibody. Overlay histogram showing MCF-7 cells stained with (Blue line). To facilitate intracellular staining, cells were fixed with 4% paraformaldehyde and permeabilized with permeabilization buffer. The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-MID2 antibody (1 ug/million cells) for 30 min at 20oC. DyLight 488 conjugated goat anti-rabbit IgG (5-10 ug/million cells) was used as secondary antibody for 30 minutes at 20oC. Isotype control antibody (Green line) was rabbit IgG (1 ug/million cells) used under the same conditions. Unlabelled sample without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.



Western blot analysis of MID2 using anti-MID2 antibody. Electrophoresis was performed on a 10% SDS-PAGE gel at 80V (Stacking gel) / 120V (Resolving gel) for 2 hours. Lane 1: human MCF-7 whole cell lysates, Lane 2: human SIHA whole cell lysates, Lane 3: human PC-3 whole cell lysates, Lane 4: human whole cell lysates, Lane 5: rat lung tissue lysates, Lane 6: rat brain tissue lysates, Lane 7: mouse lung tissue lysates, Lane 8: mouse brain tissue lysates. After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-MID2 antibody at 0.5 ug/ml overnight at 4oC, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:5000 for 1.5 hour at RT. The signal was developed using an ECL Plus Western Blotting Substrate. A specific band was detected for MID2 at approximately 83 kDa. The expected molecular weight of MID2 is ~83 kDa.

#### **Description**

MID2 antibody detects E3 ubiquitin-protein ligase MID2 (also known as Midline-2 or FXY2), a microtubule-associated E3 ligase involved in cytoskeletal organization, cell polarity, and developmental morphogenesis. Encoded by the MID2 gene on chromosome Xq22.3, this protein belongs to the tripartite motif (TRIM) family of ubiquitin ligases, featuring a RING finger domain, B-box zinc-binding motifs, a coiled-coil region, and a C-terminal COS domain that binds microtubules. MID2 functions in ubiquitination of cytoskeletal and signaling proteins, coordinating cell structure and trafficking processes crucial for morphogenesis and neurodevelopment.

In human cells, MID2 localizes along microtubules, at the centrosome, and in perinuclear regions. It associates with its close paralog MID1, with which it shares structural and functional similarities. Together, MID1 and MID2 regulate the stability of PP2A catalytic subunits and participate in tubulin network maintenance. MID2 expression is particularly high in brain, testis, and embryonic tissues, reflecting roles in neuronal migration and organ development. Mutations in MID2 have been associated with X-linked intellectual disability and developmental disorders affecting craniofacial and skeletal structures.

The MID2 antibody is widely used in cell biology and developmental research to study microtubule organization, ubiquitination, and cell morphology. Western blot analysis detects a ~78 kilodalton band corresponding to full-length MID2, while immunofluorescence reveals filamentous staining patterns overlapping with microtubules. MID2 ubiquitinates several substrates involved in cytoskeletal remodeling and signaling, including MID1-associated proteins and small GTPase regulators. Through these activities, it influences cell polarity, division, and migration. In neurons, MID2 contributes to axon guidance and growth cone dynamics.

Dysregulation of MID2 expression or mutation of its functional domains can lead to developmental abnormalities and neurological defects. Overexpression has been observed in certain cancers, where it may enhance cell motility and invasiveness through altered cytoskeletal signaling. The MID2 antibody provides a robust tool for investigating these pathways and identifying mechanisms of microtubule regulation in both normal and diseased states. NSJ Bioreagents supplies this antibody validated for its applications, supporting research into cytoskeletal dynamics, developmental disorders, and ubiquitin-dependent regulation.

### **Application Notes**

Optimal dilution of the MID2 antibody should be determined by the researcher.

#### Immunogen

A synthetic peptide corresponding to a sequence in the middle region of human MID2 was used as the immunogen for the MID2 antibody.

## Storage