

Monoclonal anti-human 14-3-3 epsilon antibody (clone AT4F8)

Mouse IgG_{2b}, κ

Cat. No. IBATGA0124

Immunogen: Recombinant human14-3-3 epsilon (1-255aa) purified from E. coli

NCBI Accession No.: NP_006752

Isotype: Mouse IgG_{2b} heavy chain and κ light chain

Clone: Anti-human 14-3-3 epsilon mAb, clone AT4F8, is derived from hybridization of mouse F0 myeloma cells with spleen cells from BALB/c mice immunized with a recombinant human 14-3-3 epsilon protein.

Description: The 14-3-3 family of proteins plays a key regulatory role in signal transduction, checkpoint control, apoptotic and nutrient-sensing pathways. 14-3-3 proteins are highly conserved and ubiquitously expressed. There are at least seven isoforms, β, γ, ε, σ, ζ, τ and η that have been identified in mammals. The 14-3-3 epsilon, a subtype of the 14-3-3 family of proteins, was thought to be brain and neuron-specific. It has been shown to interact with CDC25 phosphatases, RAF1 and IRS1 proteins, suggesting its role in diverse biochemical activities related to signal transduction, such as cell division and regulation of insulin sensitivity. It has also been implicated in the pathogenesis of small cell lung cancer.

Concentration: 1 mg/ml

Form: Liquid. In Phosphate-Buffered Saline (pH 7.4) with 0.02% Sodium Azide, 10% Glycerol

Storage: Can be stored at +4C. For long term storage, aliquot and store at -20C. Avoid repeated freezing and thawing cycles.

Usage: The antibody has been tested by ELISA, Western blot analysis, Flow cytometry and ICC/IF to assure specificity and reactivity. Since application varies, however, each investigation should be titrated by the reagent to obtain optimal results.

Application: ELISA, WB, Flow cytometry, ICC/IF



Western blot analysis

The cell lysates (40ug) were resolved by SDS-PAGE, transferred to PVDF membrane and probed with anti-human 14-3-3 epsilon antibody (1:1000). Proteins were visualized using a goat anti-mouse secondary antibody conjugated to HRP and an ECL detection system.

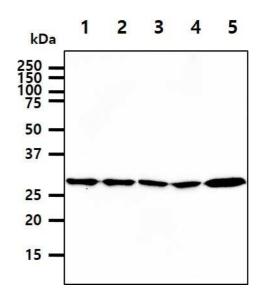
Lane 1.: HeLa cell lysate

Lane 2.: Jurkat cell lysate

Lane 3.: 293T cell lysate

Lane 4.: A549 cell lysate

Lane 5.: Mouse Brain Tissue lysate



Western blot analysis

The Recombinant Protein (50ng) were resolved by SDS-PAGE, transferred to PVDF membrane and probed with antihuman 14-3-3 epsilon antibody (1:1000). Proteins were visualized using a goat anti-mouse secondary antibody conjugated to HRP and an ECL detection system.

Lane 1.: 14-3-3 Zeta Recombinant Protein

Lane 2.: 14-3-3 Beta Recombinant Protein

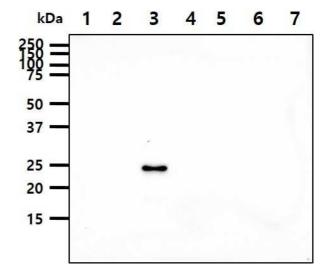
Lane 3.: 14-3-3 Epsilon Recombinant Protein

Lane 4.: 14-3-3 Eta Recombinant Protein

Lane 5.: 14-3-3 Gamma Recombinant Protein

Lane 6.: 14-3-3 Sigma Recombinant Protein

Lane 7.: 14-3-3 Tau Recombinant Protein



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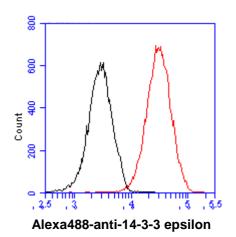
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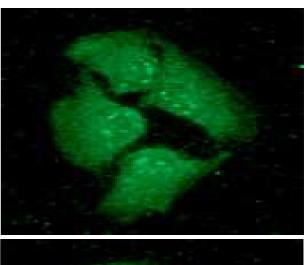
Flow cytometry

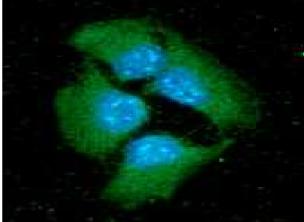
Flow cytometry analysis of 14-3-3 epsilon in HeLa cell line, staining at 2-5ug for 1x10⁶cells (red line). The secondary antibody used goat anti-mouse IgG Alexa fluor 488 conjugate. Isotype control antibody was mouse IgG (black line).



ICC/IF analysis

ICC/IF analysis of 14-3-3 epsilon in HeLa cells line, stained with DAPI (Blue) for nucleus staining and monoclonal antihuman 14-3-3 epsilon antibody (1:100) with goat anti-mouse IgG-Alexa fluor 488 conjugate (Green).

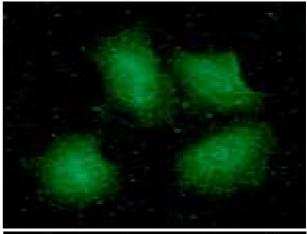


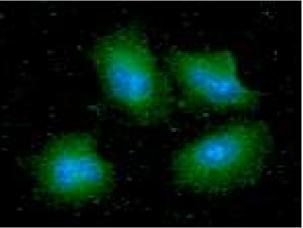




ICC/IF analysis

ICC/IF analysis of 14-3-3 epsilon in A549 cells line, stained with DAPI (Blue) for nucleus staining and monoclonal antihuman 14-3-3 epsilon antibody (1:100) with goat anti-mouse IgG-Alexa fluor 488 conjugate (Green).





General references: Oriente F., et al. (2005) J Biol Chem. 280(49): 40642-9.

Conklin D., et al. (1995) PNAS. 92(17): 7892-6.