

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human 14-3-3ζ in direct ELISAs and Western blots. Detects Mouse and Rat 14-3-3ζ in Western Blots. In direct ELISAs, no cross-reactivity with recombinant human 14-3-3 beta, theta, eta, gamma, sigma, or epsilon is observed.
Source	Monoclonal Mouse IgG _{2B} Clone # 818515
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	<i>E. coli</i> -derived recombinant human 14-3-3ζ Asp2-Asn245 Accession # P63104
Formulation	Lyophilized from a 0.2 μm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 μm filtered solution in PBS.

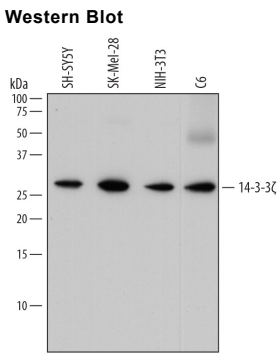
APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. *General Protocols* are available in the *Technical Information* section on our website.

	Recommended Concentration	Sample
Western Blot	0.2 μg/mL	See Below
Immunohistochemistry	8-25 μg/mL	See Below
Simple Western	2 μg/mL	See Below

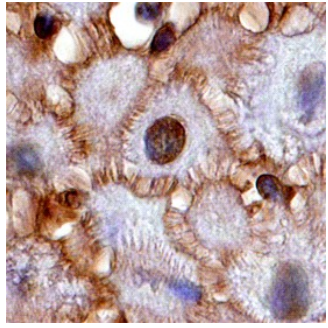
DATA

Western Blot



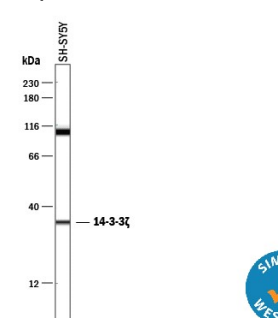
Detection of Human, Mouse, and Rat 14-3-3ζ by Western Blot. Western blot shows lysates of SH-SY5Y human neuroblastoma cell line, SK-Mel-28 human malignant melanoma cell line, NIH-3T3 mouse embryonic fibroblast cell line, and C6 rat glioma cell line. PVDF membrane was probed with 0.2 μg/mL of Mouse Anti-Human 14-3-3ζ Monoclonal Antibody (Catalog # MAB2669) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF018). A specific band was detected for 14-3-3ζ at approximately 27 kDa (as indicated). This experiment was conducted under reducing conditions and using *Immunoblot Buffer Group 1*.

Immunohistochemistry




14-3-3ζ in Human Squamous Cell Carcinoma. 14-3-3ζ was detected in immersion fixed paraffin-embedded sections of human squamous cell carcinoma using Mouse Anti-Human 14-3-3ζ Monoclonal Antibody (Catalog # MAB2669) at 15 μg/mL overnight at 4 °C. Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using Antigen Retrieval Reagent-Basic (Catalog # CTS013). Tissue was stained using the Anti-Mouse HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS002) and counterstained with hematoxylin (blue). Specific staining was localized to nuclei and plasma membrane. View our protocol for *Chromogenic IHC Staining of Paraffin-embedded Tissue Sections*.

Simple Western



Detection of Human 14-3-3ζ by Simple Western™. Simple Western lane view shows lysates of SH-SY5Y human neuroblastoma cell line, loaded at 0.5 mg/mL. A specific band was detected for 14-3-3ζ at approximately 34 kDa (as indicated) using 2 μg/mL of Mouse Anti-Human 14-3-3ζ Monoclonal Antibody (Catalog # MAB2669). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.



PREPARATION AND STORAGE

Reconstitution	Sterile PBS to a final concentration of 0.5 mg/mL.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <ul style="list-style-type: none"> ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

14-3-3 proteins are a highly conserved family of homo- and heterodimeric phosphoserine/threonine-binding proteins present in high abundance in all eukaryotic cells. 14-3-3 proteins were the first polypeptides shown to have pSer/Thr binding properties, generally recognizing the consensus sequences RSXpSXP and RXY/FXpSXP (where X is any amino acid). 14-3-3 proteins act as key regulators of intracellular signal transduction through their ability to bind specific motifs phosphorylated on serine or threonine. For example, the binding of 14-3-3 to phosphorylated BAD blocks its proapoptotic association with Bcl-XL. There are at least seven distinct 14-3-3 genes in vertebrates, alpha/beta, epsilon, gamma, theta, sigma and zeta (α/β , ϵ , η , γ , τ/θ , σ , and ζ/δ). 14-3-3 zeta, also known as Tyrosine 3-Monooxygenase/Tryptophan 5-Monooxygenase Activation Protein, zeta isoform (gene name YWHAZ) is a 245 amino acid, 27 kDa protein.