

DESCRIPTION

Species Reactivity	Human
Specificity	Detects Phospho-Paxillin (Y31) in direct ELISAs and Western blots.
Source	Monoclonal Mouse IgG _{2A} Clone # 698239
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	Phosphopeptide containing the human Paxillin Y31 site
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 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	0.1 µg/mL	See Below
Immunocytochemistry	8-25 µg/mL	See Below

DATA

Western Blot

Detection of Human Phospho-Paxillin (Y31) by Western Blot. Western blot shows lysates of A431 human epithelial carcinoma cell line, Jurkat human acute T cell leukemia cell line, and Daudi human Burkitt's lymphoma cell line untreated (-) or treated (+) with 1 mM Pervanadate (PV) for 30 minutes. PVDF membrane was probed with 0.1 µg/mL of Mouse Anti-Human Phospho-Paxillin (Y31) Antigen Affinity-purified Monoclonal Antibody (Catalog # MAB61641) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF007). Specific bands were detected for Phospho-Paxillin (Y31) at approximately 65 to 68 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunocytochemistry

Paxillin in HUVEC Human Cells. Paxillin phosphorylated at Y31 was detected in immersion fixed HUVEC human umbilical vein endothelial cells using Mouse Anti-Human Phospho-Paxillin (Y31) Antigen Affinity-purified Monoclonal Antibody (Catalog # MAB61641) at 10 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Mouse IgG Secondary Antibody (red, upper panel; Catalog # NL007) and counterstained with DAPI (blue, lower panel). Specific staining was localized to nuclei and focal adhesions. View our protocol for [Fluorescent ICC Staining of Cells on Coverslips](#).

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

Paxillin is a 65 kDa cytoskeletal adaptor protein and member of the Paxillin family. Human Paxillin is 591 amino acids (aa) in length and contains four LIM zinc-binding domains. Alternative splicing produces three isoforms. Human Paxillin shares 94% and 85% aa identity with mouse and rat Paxillin, respectively. Paxillin is found at the interface between actin filaments and the plasma membrane, and it localizes to focal adhesions, where it provides a platform for the integration and coordination of adhesion- and growth factor-related signals. Paxillin phosphorylation at tyrosines 31 and 118 is required for integrin-mediated cytoskeletal reorganization, and may play a role in the disassembly of focal adhesions and stress fibers during cellular transformation.