

DESCRIPTION

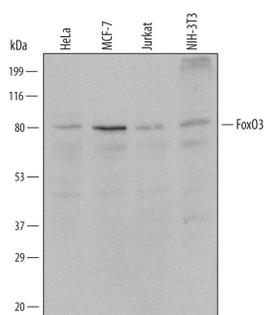
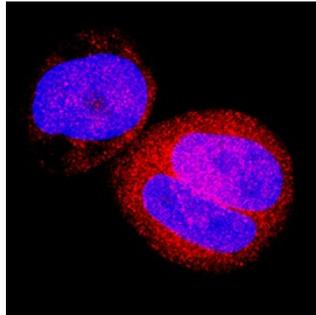
Species Reactivity	Human/Mouse
Specificity	Detects human and mouse FoxO3 in Western blots.
Source	Monoclonal Mouse IgG _{2A} Clone # 648716
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	<i>E. coli</i> -derived recombinant human FoxO3 Ala373-Gly673 Accession # O43524
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	1 µg/mL	See Below
Immunocytochemistry	5-25 µg/mL	See Below

DATA

<p>Western Blot</p>  <p>Detection of Human and Mouse FoxO3 by Western Blot. Western blot shows lysates of HeLa human cervical epithelial carcinoma cell line, MCF-7 human breast cancer cell line, Jurkat human acute T cell leukemia cell line, and NIH-3T3 mouse embryonic fibroblast cell line. PVDF Membrane was probed with 1 µg/mL of Mouse Anti-Human/Mouse FoxO3 Monoclonal Antibody (Catalog # MAB6165) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF007). A specific band was detected for FoxO3 at approximately 85 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.</p>	<p>Immunocytochemistry</p>  <p>FoxO3 in MCF-7 Human Cell Line. FoxO3 was detected in immersion fixed MCF-7 human breast cancer cell line using Mouse Anti-Human/Mouse FoxO3 Monoclonal Antibody (Catalog # MAB6165) at 8 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Mouse IgG Secondary Antibody (red; Catalog # NL007) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm. View our protocol for Fluorescent ICC Staining of Cells on Coverslips.</p>
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PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.5 mg/mL in sterile PBS.
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

Forkhead box O3 (FoxO3) is a ubiquitously expressed transcriptional regulator that is involved in cellular differentiation, angiogenesis, tumor progression, apoptosis, and the responses to oxidative stress and DNA damage. Phosphorylation of FoxO3 by Akt induces its association with 14-3-3 proteins and its retention in the cytoplasm. In response to the loss of survival factors, dephosphorylation of FoxO3 induces its translocation to the nucleus where it promotes apoptosis. Its level of acetylation is regulated in response to cellular metabolic requirements. Within amino acids 372-673 (C-terminal to the DNA binding domain), human FoxO3 shares 95% aa sequence identity with mouse and rat FoxO3.