

#2497 Store at -20°C

FoxO3a (75D8) Rabbit mAb

✓ 100 µl (10 western blots)



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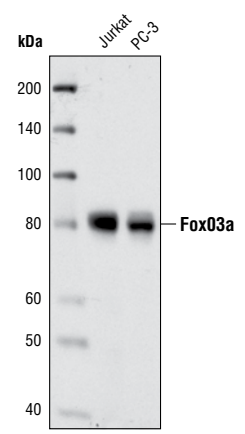
This product is intended for research purposes only. This product is not intended to be used for therapeutic or diagnostic purposes in humans or animals.

| Applications | Species Cross-Reactivity* | Molecular Wt. | Isotype |
|------------------------|---------------------------|---------------|--------------|
| W, IF-IC Endogenous | H, M, R, MK | 82-97 kDa | Rabbit IgG** |

Background: The Forkhead family of transcription factors is involved in tumorigenesis of rhabdomyosarcoma and acute leukemias (1-3). Within the family, three members (FoxO1, FoxO4 and FoxO3a) have sequence similarity to the nematode orthologue DAF-16, which mediates signaling via a pathway involving IGF1R, PI3K and Akt (4-6). Active forkhead members act as tumor suppressors by promoting cell cycle arrest and apoptosis. Increased expression of any FoxO member results in the activation of the cell cycle inhibitor p27Kip1. Forkhead transcription factors also play a part in TGF-β-mediated upregulation of p21CIP1, a process negatively regulated through PI3K (7). Increased proliferation results when forkhead transcription factors are inactivated through phosphorylation by Akt at Thr24, Ser256 and Ser319, which results in nuclear export and inhibition of transcription factor activity (8). Forkhead transcription factors can also be inhibited by the deacetylase sirtuin (SirT1) (9).

Specificity/Sensitivity: FoxO3a (75D8) Rabbit mAb detects exogenous and endogenous levels of total FoxO3a protein. The antibody does not detect the exogenously expressed family members FoxO4 or FoxO1.

Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to residues surrounding Glu50 of human FoxO3a.



Western blot analysis of extracts from Jurkat and PC-3 cells using FoxO3a (75D8) Rabbit mAb.

Entrez-Gene ID #2309
Swiss-Prot Acc. #O43524

Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at -20°C. Do not aliquot the antibody.

*Species cross-reactivity is determined by western blot.

**Anti-rabbit secondary antibodies must be used to detect this antibody.

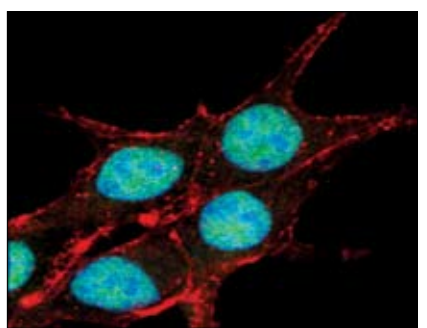
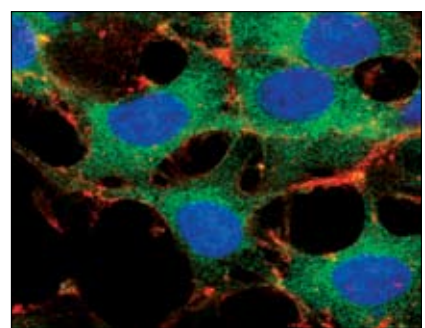
Recommended Antibody Dilutions:

| | |
|----------------------------|--------|
| Western blotting | 1:1000 |
| Immunofluorescence (IF-IC) | 1:200 |

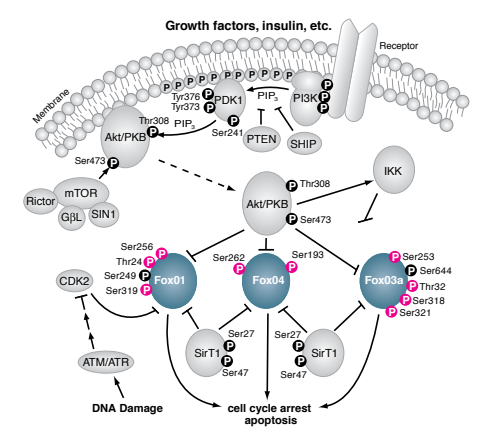
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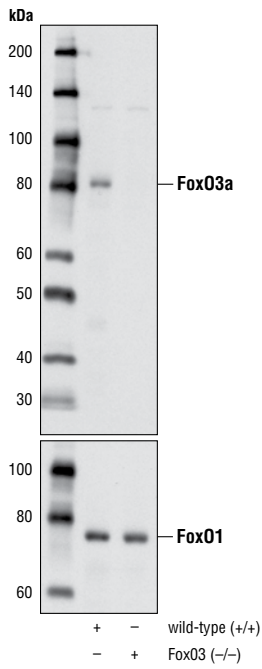
Confocal immunofluorescent analysis of SH-SY5Y cells, IGF-I treated (left) or LY294002 treated (right) using FoxO3a (75D8) Rabbit mAb (green). Actin filaments have been labeled with Alexa Fluor® 555 phalloidin (red). Blue pseudocolor = DRAQ5® #4084 (fluorescent DNA dye).



IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v BSA, 1X TBS, 0.1% Tween-20 at 4°C with gentle shaking, overnight.

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Applications Key: W—Western IP—Immunoprecipitation IHC—Immunohistochemistry ChIP—Chromatin Immunoprecipitation IF—Immunofluorescence F—Flow cytometry E-P—ELISA-Peptide
Species Cross-Reactivity Key: H—human M—mouse R—rat Hm—hamster Mk—monkey Mi—mink C—chicken Dm—D. melanogaster X—Xenopus Z—zebrafish B—bovine
Dg—dog Pg—pig Sc—S. cerevisiae Ce—C. elegans Hr—Horse All—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.



Western blot analysis of extracts from wild type (+/+) and FoxO3 knock-out (-/-) mouse brain, using FoxO3a (75D8) Rabbit mAb (upper) and FoxO1 (C29H4) Rabbit mAb #2880 (lower). (FoxO3 knock-out (-/-) mouse brain was kindly provided by Dr. Ron DePinho, Dana-Farber Cancer Institute, Boston, MA).

Background References:

- (1) Anderson, M.J. et al. (1998) *Genomics* 47, 187–199.
- (2) Galili, N. et al. (1993) *Nat. Genet.* 5, 230–235.
- (3) Borkhardt, A. et al. (1997) *Oncogene* 14, 195–202.
- (4) Nakae, J. et al. (1999) *J. Biol. Chem.* 274, 15982–15985.
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- (7) Seoane, J. et al. (2004) *Cell* 117, 211–223.
- (8) Arden, K.C. (2004) *Mol. Cell* 14, 416–418.